Microbial Effects on Ni and Cd Sorption and Transport in Volcanic Tuff

by

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I dedicate this work to my mother and deceased father who have provided love and support throughout my life.

Chapter 1

Introduction

INTRODUCTION

This thesis consists of a submitted journal article and supporting appendices. The purpose of the project was to determine the effect of microbes on cationic transition-metal sorption and transport. The project findings can be applied to land-based metal storage plans and to heavy-metal waste remediation. The methods, results, and discussion of this work are presented in the journal article titled *Microbial Effects on Ni and Cd Sorption and Transport in Volcanic Tuff*, which was submitted to the Journal of Environmental Quality in June, 1993. The objective of the study was to quantify the sorption effects of microbes on Cd^{2+} and Ni^{2+} to crushed volcanic tuff; then determine what effect the microbe which most influenced Ni^{2+} sorption had on Ni^{2+} transport in the tuff. Batch equilibrium experiments were performed with aqueous metal solutions. Transport studies were performed by pumping aqueous solution through packed columns. In both sorption and transport experiments, I compared the microbial effects in sterile media to the effects in inoculated media.

Appendix A relates the biological classification of each microbial strain studied to the identification number used to identify strains in other appendices, in my laboratory notebooks, in the frozen cultures at the NMIMT microbiology laboratory, and other persons or groups who collected or stored the strains.

Appendix B contains results from Ni^{2+} batch equilibration experiments. Eight experiments were performed to determine the effect of freshly-washed microbes on Ni^{2+} sorption to crushed volcanic tuff. Other experiments quantify the effect of NaN_3 and of freeze-dried *Bacillus megaterium* on Ni^{2+} sorption to tuff. The appendix also displays results from an experiment performed to determine sorption of Ni^{2+} to crushed tuff in a buffered system for both sterile samples and for samples inoculated with freshly-washed *B. megaterium.*

Appendix C contains the results of two batch equilibrium experiments performed to quantify Cd^{2+} sorption to the crushed volcanic tuff and the results of a kinetic study to determine when sorbed Cd^{2+} reaches an equilibrium with Cd^{2+} in solution.

Appendix D contains results from the transport study performed using four columns. The appendix includes effluent concentrations from a Br^- tracer study, Ni^{2+} effluent concentration, results from liquid scintillation counts of samples from each bottle of influent solution, and information on the effluent flux, the water volume in the apparatus, the bulk density, and the influent and effluent pH.

Appendix E contains data on microbial numbers determined by direct microscopic count and by plate counts. I performed direct microscopic cell counts on suspensions used in batch equilibration studies and plate counts on microbes in batch equilibrium samples and in media contained in the columns.

Appendix F contains information on how the autoclave affects the moisture content of tuff.

Chapter 2

Paper Titled "Effect of Microbes on Ni and Cd Sorption and Transport in Volcanic Tuff"

by R. L. Brown, R. S. Bowman, and T. L. Kieft

ABSTRACT

We performed sorption and transport experiments to determine how vadose-zone microbes affect sorption of Ni^{2+} and Cd^{2+} on crushed volcanic tuff and transport of Ni^{2+} in the same medium. Sorption of Ni^{2+} and Cd^{2+} to the tuff was less in samples inoculated with microbes than in sterile samples. Since Ni sorption was the same both in the presence and absence of microbes when a buffer was used, microbes appear to decrease sorption by decreasing solution pH. Samples with *Bacillus megaterium* decreased Ni^{2+} sorption the most of the 14 subsurface microbial strains tested. Nickel isotherms were linear up to initial concentrations of 10 mg L^{-1} with an average distribution coefficient (k_d) of 7.4 L kg^{-1} for samples with freeze-dried *B. megaterium* and 50 L kg^{-1} for sterile samples containing 1 g L^{-1} NaN_3. Sterile samples without NaN_3 had a higher k_d (184 to 265 L kg^{-1}). We therefore predicted that the retardation of Ni in inoculated columns would be lower than in sterile columns. A transport experiment with both inoculated (inoculum consisted of freeze-dried *B. megaterium*) and sterile (solutions contained NaN_3) columns indicated that retardation (R) of Ni was less for columns with microbes (R = 205) than for sterile columns (R = 307), but retardation was higher than predicted by batch sorption experiments in both groups of columns. Dispersion was lower in inoculated columns, possibly due to microbial clogging of secondary pores. The results indicate that microbes can increase the mobility of cationic metals in subsurface environments.

INTRODUCTION

Preventing hazardous wastes from reaching sensitive environments and remediating waste that is already in the environment requires understanding of microbiological effects on hazardous waste fate and transport. Microbes may significantly influence metal contaminant transport by either increasing or decreasing sorption of metals to surfaces in porous media. Francis (1990) summarized microbial influences on metal solubility as (1) changes in the pH of the solution, (2) redox reactions which affect metal valence states and solubility, (3) chelation, solubilization, and leaching by microbial metabolites and decomposition products, (4) biomethylation and production of volatile and/or toxic alkylated metal compounds, and (5) biodegradation of metal-organic complexes. Microbes are also capable of sorbing metals (Kelly et al., 1979; Beveridge, 1986). This could lead to increased proportions of metals in the solid phase, but also to increased colloidal transport of the metals. These effects need to be considered for remediation efforts and for prediction of contaminant fate.

Several studies have shown that microbes can decrease sorption of Cd^{2+} and Ni^{2+} to a medium. Chanmugathas and Bollag (1987) determined that mobilization of strongly bound Cd is microbially mediated. For some but not all soils in their study, the release of Cd was accompanied by a decrease in the soil suspension pH. Burke et al. (1991) determined that sediments containing microbes sorb less Cd than sediments that are autoclaved. In a study by Gerringa (1990) many metals, including Cd, increased in mobility concomitant with aerobic degradation of organic matter. Wildung et al. (1979) found that many metal-tolerant microbes produce metabolites that complex with

Ni and increase Ni transport through soil.

The objective of this study was to determine the effects of vadose-zone microbes on the sorption of Ni^{2+} and Cd^{2+} and transport of Ni^{2+} in crushed volcanic tuff. The tuff we used is similar to that which underlies the proposed high-level radioactive waste repository at Yucca Mountain, Nevada, and low-level radioactive waste disposal sites at Los Alamos National Laboratory, New Mexico. Both Ni and Cd, as well as other heavy metals, may occur in industrial waste, in sewage sludge, and in long-term waste storage facilities. Ni^{2+} served as a model of other heavy metals in the +2 valence state. Ni^{2+} is stable in aqueous solutions over a wide range of Eh and pH (Deltombe et al., 1974). Although high Ni concentrations are toxic (Drake, 1988), Ni is an essential trace element for bacteria, plants, and animals (Ankel-Fuchs and Thauer, 1988). Many bacteria require Ni for the synthesis of nickel hydrogenase. Nickel toxicity in microorganisms depends on the type of organism and on the growth medium (Kaltwasser and Frings,1980). We performed sorption experiments with Cd^{2+} for comparison with the Ni data. Cd has been implicated in health problems in humans and animals (Friberg et al., 1971).

MATERIALS AND METHODS

Porous Medium

The porous medium was Bandelier tuff which was crushed and sieved to less than 250 µm. The tuff was obtained from a rock quarry along Jemez Road across from the Meson Center SE of the city of Los Alamos, NM. Twenty-nine percent of the crushed tuff was between 149 and 250 µm, 49% was between 44 and 149 µm and 22% was less than 44 µm in diameter. X-ray fluorescence analysis indicated that the tuff was composed primarily of SiO_2, with significant portions of Al, K, Na, and Fe oxides (Table 1). X-ray diffraction analysis indicated that the tuff contained much quartz, minor anorthoclase, and either cristobalite or opal, but no clay minerals.

Microbes

We investigated sorption effects of 14 microbial strains that were isolated from the vadose zone of the Pajarito Plateau at Los Alamos National Laboratory (LANL) in July, 1987. The microbes were present in tuffaceous cores collected using a continuous auger drilling technique and sterile split-spoon samplers (Hersman et al., 1988). Depths of sampling ranged from 15.2 to 53.3 m. The core samples contained 10 to 100 culturable microbes or colony forming units (CFU) g^{-1} dry weight soil and 2.0 to 9.5 x 10^6 total cells g^{-1} based on direct microscopic counts. Microbial cultures were supplied by W. C. Ghiorse, Cornell University. To preserve the cultures, a mixture consisting of 50% glycerol and 50% cells suspended in a complex growth medium was frozen at -80°C. The microbes, identified by fatty-acid methyl ester profiles (Microbial ID, Inc., Newark, DE), consisted of yeasts and both Gram-negative and Gram-positive

Table 1. Composition of tuff as determined by X-Ray fluorescence.

Oxide	Weight %
SiO_2	79.5
TiO_2	0.09
Al_2O_3	11.9
Fe_2O_3-T*	1.37
MgO	0.04
CaO	0.15
K_2O	3.97
Na_2O	3.97
MnO	0.05
P_2O_5	0.01
S	0.007
LOI**	0.45
Total	100.51

*Fe_2O_3-T = Total iron calculated as Fe_2O_3

**LOI = Loss on ignition

bacillus and coccoid bacteria (Table 2). One unidentified yeast from 15.2 m had a profile that was similar to both *Cryptococcus* and *Candida*.

Sorption Experiments

Prior to the batch equilibration experiments, we acid-washed 250-mL polypropylene flat-bottom centrifuge bottles and 50-mL polyallomer Oak Ridge-Type centrifuge tubes in 5% HCl solution. We made all solutions using NANOpure water (Barnstead, Dubuque, IA). All solutions, centrifuge bottles (used for washing microbes), and glass pipettes were sterilized by autoclaving at 121°C for 20 min. To prepare the sterile tuff, we first measured 10 g of the crushed tuff into the centrifuge tubes, then autoclaved the tubes for 1 h at 121°C for three successive days. To produce sufficient quantities of microbes for the experiments, we inoculated a broth consisting of 1.0 g glucose, 1.0 g Difco yeast extract (Difco Laboratories, Detroit, MI), 0.5 g Difco bactopeptone, 0.6 g $MgSO_4 \cdot H_20$, and 0.07 g $CaCl_2 \cdot 2H_20$ in 1 L distilled water. This is a modification of a growth medium developed by Balkwill and Ghiorse (1985). The incubating microbes were shaken on a rotary shaker at 150 rpm at room temperature (about 25°C) for two days. We washed the cells by centrifuging at 19000 x g, pouring off the supernatant, and resuspending the pellet in 0.005 M $CaCl_2$ solution. The washing procedure was repeated twice. The optical density of the final suspension averaged 0.76 with a standard deviation (sd) of 0.18) for all experiments performed with freshly washed microbes.

We performed all batch equilibrium experiments in triplicate. We mixed 10 g of sterile tuff with 10 mL of 0.005 M $CaCl_2$ solution for sterile samples or 10 mL of the microbial suspension for inoculated samples. Each sample was vortex-mixed for 30 s and then placed on a reciprocal shaker at 100 rpm for about 16 h, allowing the microbes to adjust to new conditions. We then added

Table 2. Microbes identified by fatty-acid methyl ester profiles and depths from which they were obtained.

Classification		Depth (m)	# of strains
Bacteria:	*Arthrobacter oxydans*	45.7	1
		53.3	3
	Bacillus megaterium	53.3	1
	Clavibacter michiganense insidiosum	15.2	1
	Pseudomonas (species not known)	15.2	4
	Rhodococcus fascians	15.2	1
Yeast:	*Candida famata*	53.3	1
	Cryptococcus albidus	15.2	1
	Unidentified yeast	15.2	1

10 mL of $NiCl_2$ spiked with ^{63}Ni or 10 mL $CdCl_2$ solution at double the desired initial concentrations. We used initial N^{2+} concentrations of 0.01 to 20 mg L^{-1} and initial Cd^{2+} concentrations from 5 to 125 mg L^{-1}. To determine the relative effects of each microbial strain on Ni sorption, we used initial concentrations of 10 mg L^{-1}. All solutions were added under a HEPA filter sterile hood (Environmental Air Control, Inc., Hagerstown, MD). The samples were shaken for an additional 24 h (determined in preliminary Cd^{2+} experiments to be sufficient time to attain sorption equilibrium). Then we centrifuged them at 27000 x g for 15 min and analyzed the supernatant for metal concentrations. We used inductively coupled plasma emmission spectroscopy to analyze Cd. We quantified ^{63}Ni concentration using a Packard Tri-Carb 460 CD liquid scintillation counter (Packard Instrument Co., Downers Grove, IL). The samples for most of the Ni^{2+} batch experiments were prepared by adding 1 mL of aqueous sample to 20 mL of Universol scintilation cocktail (ICN Biomedical, Irvine, CA). The samples for the transport study and the latter batch studies were prepared by adding 1mL of aqueous sample to 14 mL of either Universol or or ScintiVerse I scintilation cocktail (Fisher Scientific, Pittsburgh, PA). There was no statistically significant difference between the counts from the two cocktail types. The quench factor was the same for all samples. We subtracted the background counts from all sample counts.

We performed other batch experiments to determine the effects of (1) using freeze-dried *Bacillus megaterium* instead of freshly grown microbes, (2) pH, and (3) NaN_3 on Ni^{2+} sorption to crushed tuff. All of these experiments were performed concurrently with sterile samples not containing NaN_3. To determine if the microbes decreased sorption by changing the solution pH, some isotherms were prepared in buffered systems. The buffer consisted of 0.57 g KH_2PO_4 and 6.24 g K_2HPO_4 mixed with 100 mL of NANOpure water to make a 0.4 M solution. We did not use $CaCl_2$ as an electrolyte in samples with buffer because a precipitate formed when $CaCl_2$ and K_2HPO_4 were mixed together. We first added 5 mL of buffered solution and 5 mL of suspension for the inoculated group of samples or 5 mL of buffered solution and 5 mL of distilled water for the sterile group of samples to 10 g of the tuff. We used an initial Ni^{2+} concentration of 10 mg L^{-1}. At the end of the experiment, the pH of the supernatant was measured using a gel-filled polymer-body combination pH electrode with an Ag/AgCl reference. To determine the NaN_3 effect on Ni^{2+} sorption, we added a 10-mL mixture of 2 g L^{-1} NaN_3 and 0.005 M $CaCl_2$ solution to 10 g of tuff. Brock (1978) showed that 1 g L^{-1} NaN_3 (the final solution concentration in our experiments) effectively inhibits microbial activity. To determine the effect of freeze-dried *B. megaterium* on Ni^{2+} sorption, we first froze a suspension of washed microbes in 0.005 M $CaCl_2$ solution at -50°C in a shell freezer (Labconco, Kansas City, MO) and dried them with a lyophilizer (Labconco). Prior to performing experiments, the freeze-dried cells were suspended in a 0.005 M $CaCl_2$ solution. Mixing the suspension with the tuff resulted in about 2.4 mg of freeze-dried microbes per g of tuff or 5.2×10^8 cells g^{-1} determined by direct count. The initial Ni^{2+} concentrations for experiments with both freeze-dried microbes and with NaN_3 were 1, 4, 7, and 10 mg L^{-1}.

Transport Experiment

The transport experiments were performed in 5-cm tall by 5-cm diameter Plexiglas columns. These columns were made from 30-cm columns purchased from Soil Measurement Systems (Tucson, AZ). The stainless steel porous endplates had air entry values of 25 kPa and pore diameters of 0.5 µm. Rubber O-rings were placed next to the porous endplates for sealing purposes. In addition we used 3.2-mm I.D. silicone tubing, polypropylene dual check valves (Manostat, New York, NY), 3-mL plastic luer-lok tip syringes, low-density polyethylene connectors, and glass influent bottles and sampling tubes.

Prior to the transport experiment, we sterilized all apparatus, solutions, and crushed tuff. We sterilized the columns, connectors, check valves, and O-rings by placing them over boiling water for 10 min on three successive days (tyndallization). Solutions, tubing, and other labware were autoclaved at 121°C for 20 min. The tuff was divided into two containers, each with 400 g, then autoclaved at 121°C for 1 h for 3 consecutive days. We dried the sterile tuff at 100°C for 24 h, then cooled it to room temperature (~25°C). Packing was done in the sterile hood using sterile gloves. The freeze-dried microbes were mixed thoroughly with one container of tuff (1.3 mg freeze-dried microbes g^{-1} tuff) prior to packing the columns. Two columns were packed with sterile tuff and two with inoculated tuff. To pack the columns, we transferred the tuff into a funnel with the bottom of the funnel placed directly on the bottom of the column. We lifted the funnel slightly so the tuff slid into the column, then moved the funnel around the bottom to distribute the tuff. After about 0.5 cm of tuff had accumulated, the column was vibrated for about 10 s to settle the tuff. Then the process was repeated until the column was full. This resulted in an average dry bulk density of 1390 kg m^{-3} for the four columns. When the packing was complete, we cleared the top of the column using a sterile pipette attached to a vacuum, then inserted the sterile rubber O-ring and placed the end plate and top on the column. The average saturated hydraulic conductivity was 2.3×10^{-6} m s^{-1} (sd = 7.1×10^{-7}) for two columns packed in this manner, determined using the falling-head permeameter method (Todd, 1959).

After packing, we filled the columns from the bottom with sterile 0.005 M $CaCl_2$ solution at a rate of 605 mL/ day. Solutions were pumped into the columns using a multichannel syringe pump (Soil Measurement Systems). To maintain sterile conditions, all solutions entering the sterile columns contained 1 g L^{-1} NaN_3. Approximately 30 pore volumes (PVs) were pumped through each column prior to adding a constant supply of 0.005 M $CaCl_2$, 9.3 mg L^{-1} Ni^{2+} solution. After about 270 PVs (~11 L) of Ni solution had passed through each column, we pumped a solution containing 8.5 mg L^{-1} Br^- (sterile columns) or 7.45 mg L^{-1} Br^- (inoculated columns) in addition to 9.3 mg L^{-1} Ni^{2+} for about 30 PVs to determine the characteristics of nonreactive tracer flow through the columns. Then we continued adding Ni solution without Br. Br^- was analyzed using high performance liquid chromotography. The total volume of Ni solution pumped was about 530 PVs (~21 L) for the sterile columns and 850 PV (~34 L) for inoculated columns. Column pore volumes were determined from the weight of water present in each column at the end of the experiment and corrected for the volume of water in the apparatus (endplates and connecting tubing).

Microbe Enumeration

To determine the number of culturable bacteria, we performed plate counts as described by Kieft and Rosacker (1991). We used growth medium consisting of 0.1 g glucose, 0.1 g Difco yeast extract, 0.05 g Difco bactopeptone, 0.6 g $MgSO_4 \cdot H_20$, 0.07 g $CaCl_2 \cdot 2H_20$, 15 g Difco Bacto-agar, and 1 L distilled water (another modification of the medium developed by Balkwill and Ghiorse, 1985). To determine how Ni^{2+} affected the viability of microbes in the saturated crushed tuff during the batch equilibrium experiment, we performed plate counts for batch equilibration experiments with both freshly cultured microbes and freeze-dried microbes on the microbe suspension, the tuff-suspension mixture immediately prior to adding Ni solution, and the tuff-suspension mixture after adding Ni and shaking for 24 h. We also performed plate counts on the crushed tuff used for column experiments. We sampled both the tuff used to pack the columns and the tuff taken from the center of each column at the end of the transport experiment. We performed direct counts using a Petroff-Hausser counting chamber (Hausser Scientific Partnership, Horsham, PA) and phase contrast microscopy to determine the number of cells in relation to culturable cells. When the samples could not be counted immediately, we preserved the 10 mL suspension with 200 µL of glutaraldehyde.

RESULTS AND DISCUSSION

Microbial Numbers

The number of culturable microbes remained approximately 10^6 to 10^7 CFU g^{-1} tuff during batch experiments using either freshly grown or freeze-dried *B. megaterium* (Table 3). We conclude that *B. megaterium* mixed with crushed tuff and aqueous solution is not susceptible to the toxic affects of Ni^{2+} for the concentration range of 0 to 100 mg L^{-1} and that the microbes continue to be metabolically active in the presence of Ni^{2+} and crushed tuff.

The dry tuff used to pack the columns contained 1.5×10^3 CFU g^{-1} tuff. After unpacking the columns, one inoculated column contained 1.0×10^5 CFU g^{-1} tuff and the other contained 1.3×10^5 CFU g^{-1}. No culturable cells were detected in the sterile columns at the end of the experiment. Assuming that the number of cells was 20 x the cultured number (as indicated by comparing plate counts of the microbial suspensions to direct counts), the volume of each microbe was 1 μm^3, and the bulk density of the tuff was 1390 kg m^{-3}, the volume occupied by the microbes in the inoculated columns was 3.2 cm^3 m^{-3} of porous medium. Assuming the area of each microbe is 0.7 μm^2, the surface area covered was 2.2 mm^2 cm^{-3}.

Ni Sorption Experiments

We prepared sorption isotherms with initial Ni concentrations of 1, 4, 7, and 10 mg L^{-1} to determine the effect of NaN_3 and of freeze-dried *B.*

Table 3. Culturable microbe numbers from batch sorption experiments using *B. megaterium*. Each sample was taken from a separate tube.

Stage sample was taken	Freshly Washed Microbes (CFU g^{-1} tuff)	Lyophilized Microbes (CFU g^{-1} tuff)
Immediately following inoculation:	4.7×10^7	2.9×10^7
After shaking slurry for 16 hours, but prior to adding Ni ion:	1.1×10^7	1.1×10^7
After adding Ni ion and shaking an additional 24 hours:		
Ni^{2+} concentration = 0 mg L^{-1}	7.9×10^6	-
Ni^{2+} concentration = 0.01 mg L^{-1}	9.3×10^6	-
Ni^{2+} concentration = 1.0 mg L^{-1}	8.0×10^7	-
Ni^{2+} concentration = 10.0 mg L^{-1}	-	1.4×10^7
Ni^{2+} concentration = 100 mg L^{-1}	1.3×10^7	-

megaterium on Ni sorption (Fig. 1). For this concentration range, the isotherms were linear and could be characterized by a distribution coefficient (k_d) where:

$$S = k_d C$$

S is the sorbed concentration and C is the solution concentration. The average k_d for sterile samples without NaN_3 was 158 L kg^{-1} (sd = 50 L kg^{-1}, the number of samples (n) = 10). Sodium azide decreased Ni^{2+} sorption (k_d = 49.9 L kg^{-1}, sd = 1.7, n = 2); however sorption in the presence of NaN_3 was significantly higher than sorption in two experiments using samples inoculated with freeze-dried *B. megaterium* (k_d = 7.4 L kg^{-1}, sd = 0.5). A factor that may lend support to these findings is that the Ni^{2+} sorption for samples inoculated with *B. megaterium* was similar to sorption of Ni^{2+} to nonsterile crushed tuff (k_d = 8.2 L kg^{-1}) in a study by Bowman et al. (1981). However, the tuff they used probably had different chemical and physical properties since it was not autoclaved and consisted of larger diameter grains.

Isotherms were also linear for initial Ni^{2+} concentrations less than 10 mg L^{-1} for other microbes including *Candida famata*, *Rhodococcus fascians*, and an *Arthrobacter oxydans* strain. We thus assumed the isotherms were linear for all microbes tested at an initial concentration of 10 mg L^{-1}. All of the microbes caused decreased sorption of Ni^{2+}; however, the degree of the effect varied among microorganisms (Fig. 2). *B. megaterium* consistently decreased sorption of Ni^{2+} more than other microorganisms. For experiments with an initial Ni^{2+} concentration of 10 mg L^{-1}, the average k_d for all 14 microbial strains tested

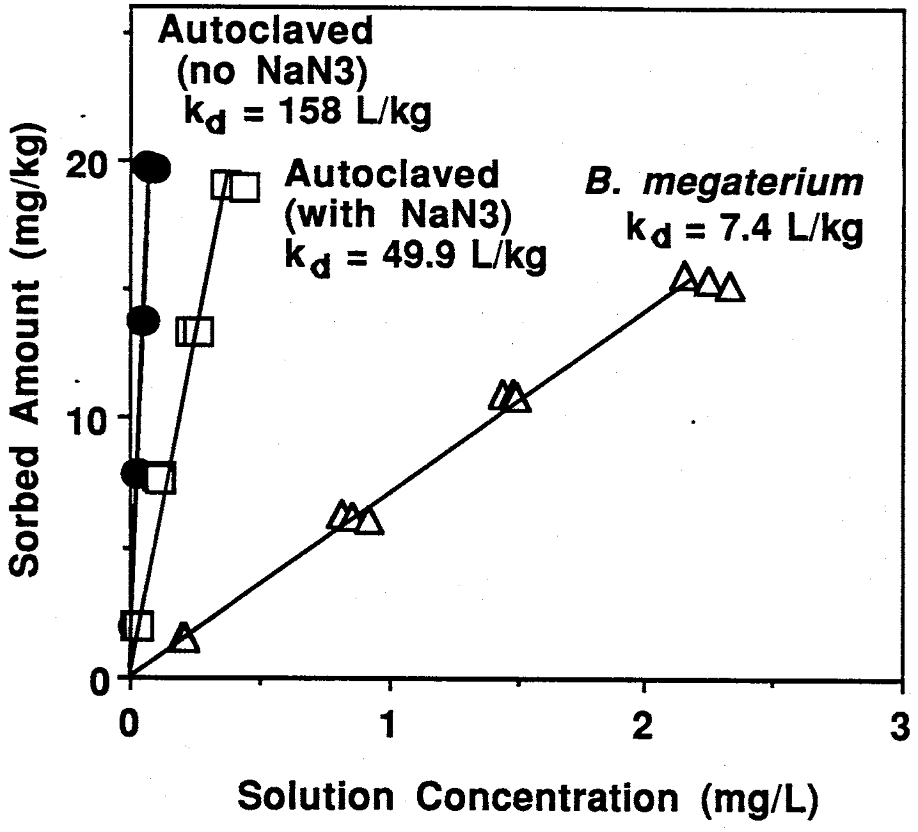

Fig. 1. Linear Ni isotherms for autoclaved tuff with and without NaN_3, and for tuff inoculated with freeze-dried *B. megaterium.*

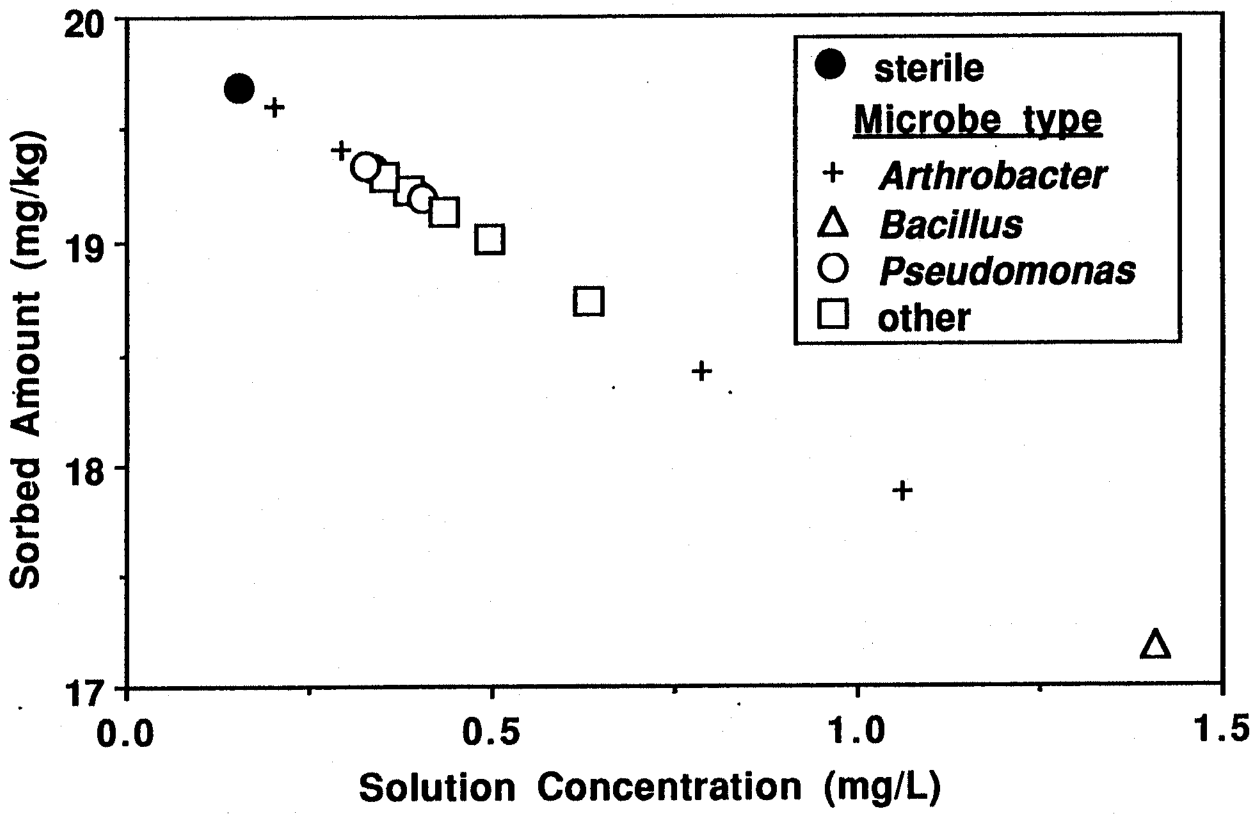

Fig. 2. The effect of each microbial strain on Ni sorption to tuff. Initial Ni^{2+} concentration was 10 mg L^{-1}. Data points are averages of experiments with each microbial strain. For most microbes, the results are from averages of triplicates from one experiment. For *B. megaterium* and *C. famata*, the results are from an average of three experiments.

was 44.5 L kg^{-1} (sd = 22.7, n = 14). All *Pseudomonas* strains affected sorption to a similar degree with an average k_d of 52.1 L kg^{-1} (sd = 5.7, n = 4); however, different strains of *Arthrobacter* exhibited varying sorption with an average k_d of 52.0 L kg^{-1}, but a standard deviation of 35.4 (n = 4). Inconsistencies between isotherms from different experiments using either sterile samples or samples inoculated with a specific microbial strain may be explained by the bacteria being in different growth phases, by different quantities of microbes being added to the sample, or by different temperatures for different experiments. Temperatures ranged from 24 to 30°C.

Although bacteria may decrease sorption of metals by decreasing the pH, producing chelating agents, or by influencing the oxidation state of the metals through redox reactions, the decreased sorption of Ni^{2+} to the crushed tuff in our experiments appears to be caused mostly by microbially produced acid (Fig. 3). For an initial concentration of 10 mg L^{-1}, supernatant pH was 1.1 units less in inoculated samples than in sterile samples. For samples buffered to pH 7.8 - 7.9, the Ni^{2+} sorption was equal in sterile samples and samples inoculated with *B. megaterium*. The supernatant resulting from washing the microbes did not affect sorption.

Cd Sorption Experiments

To determine if microbes had a similar effect on sorption of other divalent transition metals, we prepared Cd^{2+} isotherms using an initial concentration range of 6 to 126 mg L^{-1} (Figs. 4 and 5). The isotherms were logarithmic as has been previously observed in studies with Ni^{2+} and Cd^{2+} sorption on soils for

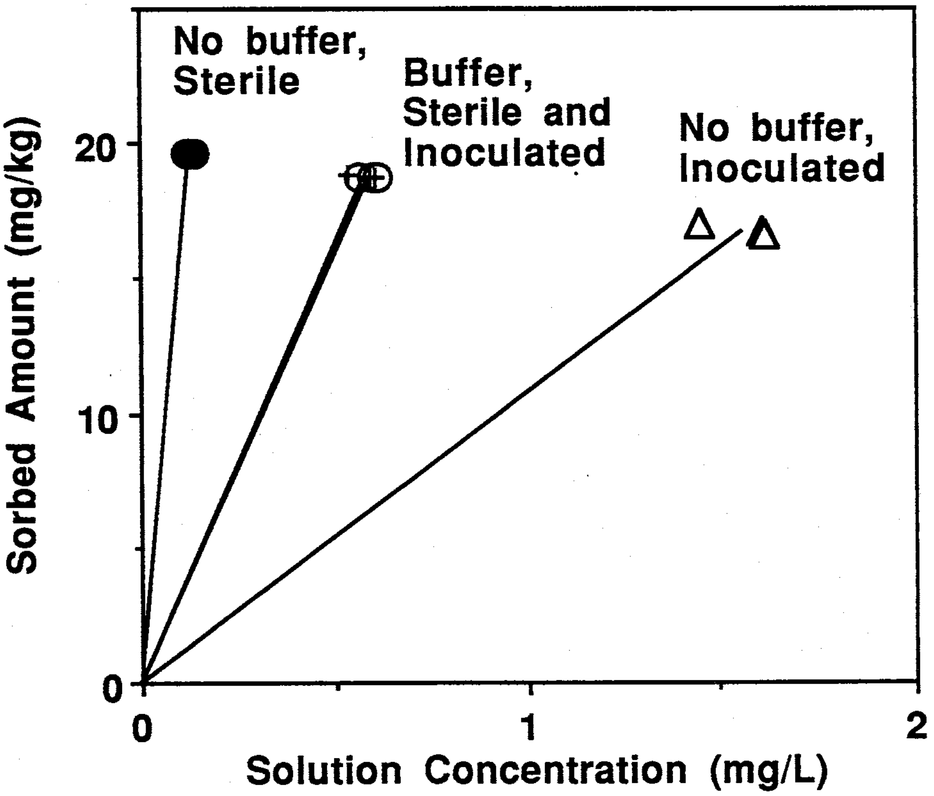

Fig. 3. Ni sorption isotherms showing the pH effect on samples inoculated with *B. megaterium*. ● = sterile samples without buffer (pH = 8.2), ○ = sterile samples with buffer (pH = 7.9), + = inoculated samples with buffer (pH = 7.8), Δ = inoculated samples without buffer (pH = 7.1)

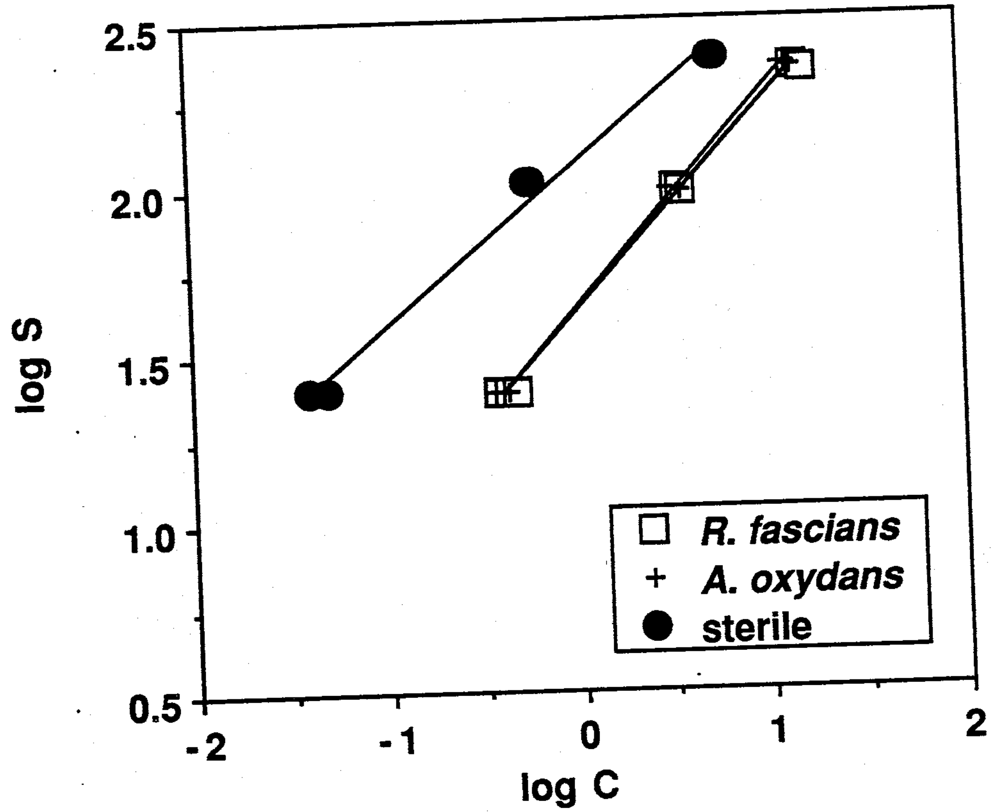

Fig. 4. Logarithmic Cd sorption isotherms for initial Cd concentrations of 13 to 120 mg L^{-1} on sterile and inoculated tuff.

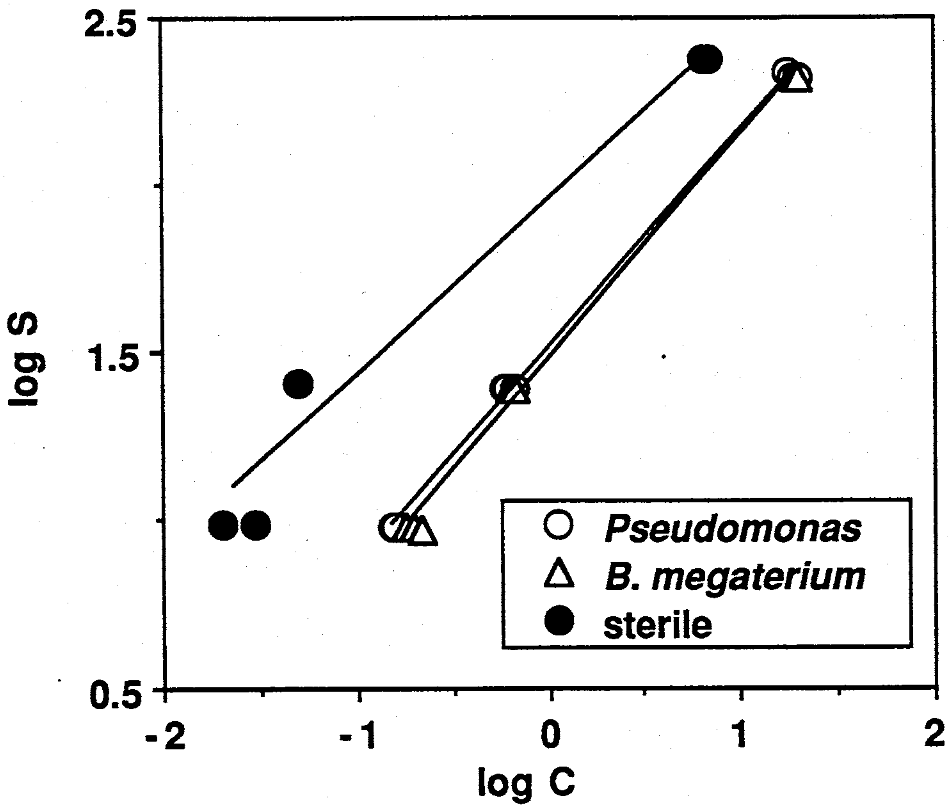

Fig. 5. Logarithmic Cd sorption isotherms for initial Cd^{2+} concentrations of 13 to 120 mg L^{-1} on sterile and inoculated tuff.

initial concentrations above 10 mg L^{-1} (Bowman et al., 1981; O'Connor et al., 1984) and can be described for both metals by the Freundlich sorption equation:

$$\log S = \log k_f + N \log C$$

where N is the Freundlich isotherm exponent and k_f is the Freundlich isotherm constant. If S is expressed in units of mg kg^{-1} and C is in units of mg L^{-1}, then N for sterile samples ranged from 0.52 and 0.62 and k_f ranged from 89 to 115. Variability may have been caused by performing experiments at different temperatures. For isotherms resulting from inoculating samples with *R. fascians*, an *A. oxydans* strain, *B. megaterium*, or a *Pseudomonas* strain, N was similar to the values for sterile treatments, ranging from 0.62 to 0.66; k_f was lower, ranging from 30 to 43. For all experiments, sorption of Cd^{2+} to tuff decreased when microbes were present. The difference in sorption may have been caused by the microbes lowering the pH of the solution; however, we did not measure the supernatant pH or perform any Cd^{2+} experiments with buffer.

Transport Experiment

Br^- and Ni^{2+} BTCs are shown in Figs. 6 and 7. We used the curve-fitting program CXTFIT (Parker and van Genuchten, 1984) to fit the parameters of the 1-dimensional advection-dispersion equation to the experimental BTCs. Assuming that R is unity for Br^- BTCs, we fitted the pore-water velocity (v), the dispersion coefficient (D), and the pulse to all four columns in the time domain, using the one-region, flux concentration model (Table 4). We used the fitted parameters from the Br^- BTCs to calculate R for Ni^{2+} BTCs (Table 5).

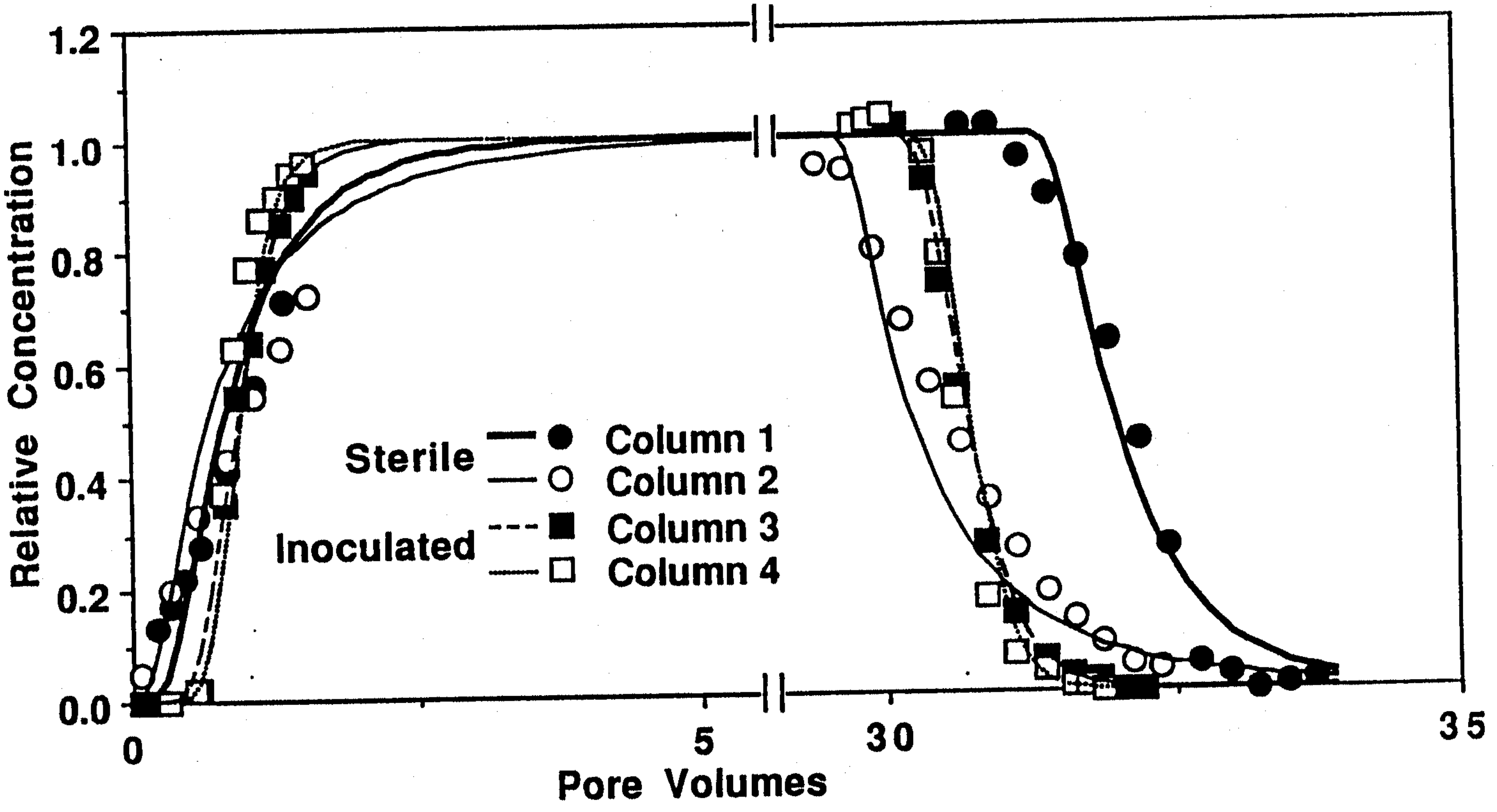

Fig. 6. Br⁻ breakthrough curves. We did not measure Br⁻ concentrations of sample effluent between 2 and 22 pore volumes. Curves were fitted using CXTFIT.

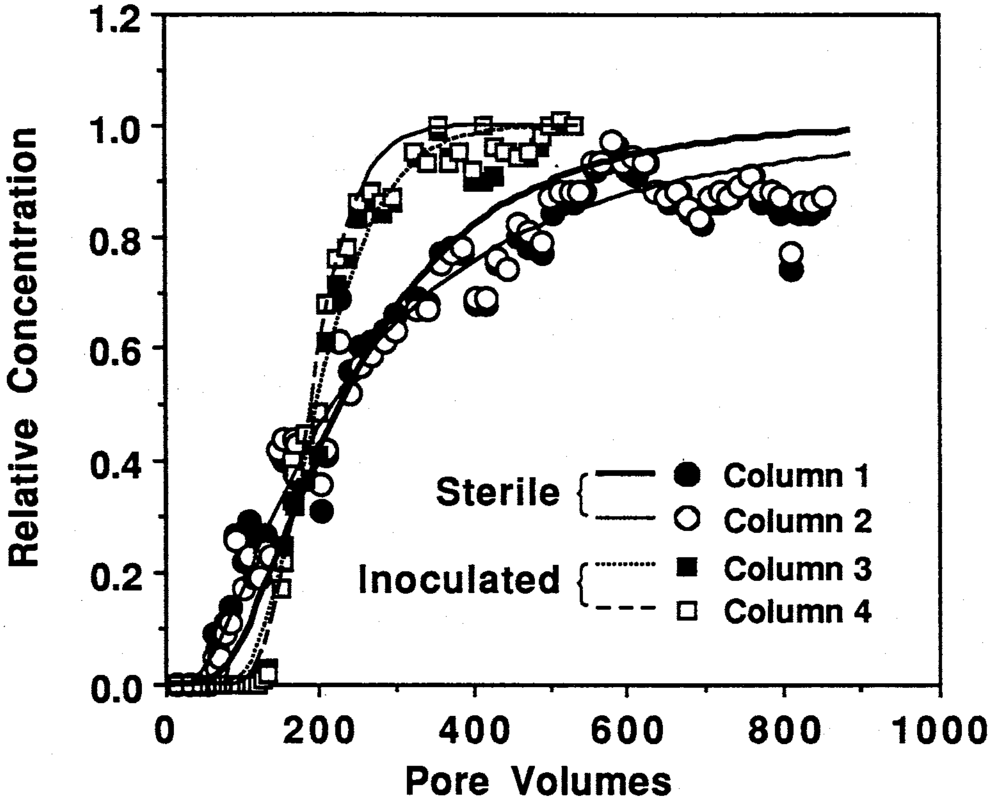

Fig. 7. Ni^{2+} breakthrough curves. Curves were fitted using CXTFIT.

Table 4. CXTFIT parameters for Br^- BTCs (one-region model). 95% confidence intervals are given.

	Sterile Columns		Inoculated Columns	
Column number	1	2	3	4
Input parameter:				
R	1	1	1	1
Fitted parameters:				
v_{Br} (cm day^{-1})	73.4 ± 10.9	56.5 ± 7.6	77.7 ± 2.5	84.8 ± 3.1
D (cm^2 day^{-1})	78.8 ± 34.2	133.7 ± 47.5	23.2 ± 4.8	14.3 ± 4.3
Pulse (day)	2.03 ± 0.01	1.99 ± 0.01	2.01 ± 0.003	1.99 ± 0.003
R^2	0.96	0.97	0.99	0.99

Table 5. CXTFIT parameters for Ni^{2+} BTCs (one region model). 95% confidence intervals are given.

	Sterile Columns		Inoculated Columns	
Column number	1	2	3	4
Input parameters:				
v_{Ni} (cm day^{-1})	61.1	48.8	66.8	71.1
D (cm^2 day^{-1})	65.6	115	19.9	12.0
Fitted parameter:				
R	276 ± 18	318 ± 14	210 ± 5	195 ± 4
R^2	0.90	0.97	0.99	0.99

Differences in water flow rates for the Br^- and Ni^{2+} BTCs were accounted for by assuming a linear relationship between velocity and flow rate. Also, D was calculated for the Ni^{2+} BTCs assuming a linear relationship between D and v (van Genuchten and Wierenga, 1986)

Using the the average k_d from the experiments with NaN_3 for sterile samples and with freeze-dried *B. megaterium* for inoculated samples, we predicted that for sterile samples, the retardation factor (R) would be 170 and for inoculated samples R would be 25, assuming that:

$$R = 1 + \frac{\rho k_d}{\theta}$$

where ρ is the bulk density of the columns and θ is the porosity of the saturated column. The average ρ of the four columns was 1390 kg m^{-3} (sd = 0.01) and the average θ was 0.405 (sd = 0.05).

The optimized values for R (Table 5) indicate that the retardation in inoculated columns was less than in sterile columns; however, all values for R are much higher than predicted from the batch isotherm data. The reasons for the greater sorption are not certain. One possible explanation is that in the dynamic system the tuff buffers the solution to a higher pH causing higher sorption. This is not supported by measurements taken of the column effluent pH because the average pH of the column effluent was between the pH of the sterile and inoculated samples in batch experiments. The pH of the sterile column effluent was 7.65 (sd = 0.35, n = 12) and of the inoculated columns was 7.77 (sd = 0.17, n = 8). This compares with the pH measurements of the nonbuffered samples of 8.18 (sd = 0.08, n = 3) for sterile samples and 7.09 (sd = 0.04, n = 3) for the inoculated samples for initial Ni^{2+} concentrations of 10 mg L^{-1}. The pH of the influent solution was 7.2. The lack of microbial effect on the pH of the columns may have been caused by the flux of solution through the material, and also there may not have been enough microbes to acidify the solution. Inoculated samples in the batch equilibrium experiments contained approximately 100 x more CFU g^{-1} than the media used for the column experiments.

The difference in R between the two sets of columns may have been caused by differences in the physical properties instead of by microbially produced acid. For example, if bacteria clogged secondary pores and if some sorption sites were located in the clogged secondary pores, then retardation would have been less in inoculated media. Since *B. megaterium* forms flocs when grown in liquid culture, it may also have formed flocs within the porous medium and thus clogged pores that were bigger than the actual size of the bacteria. Another factor suggesting that secondary pores may be present in sterile columns and not in inoculated columns is that dispersion of both Ni^{2+} and Br^- BTCs was much greater in the sterile columns than in inoculated columns. Since the values of D within each treatment were similar, we conclude that the method used in packing did not cause the dispersion difference. We believe that microbial blockage of pores may be the main explanation for decreased D and may also explain increased R; however, other factors are probably involved. One argument against secondary porosity explaining the differences in R and D is that the surface area covered by the microbes appears to be insignificant. Perhaps the surface area per microbe was greater than assumed; or alternatively, microbes may have clogged pores by producing extracellular polymeric substances.

Overall Significance

We have determined that microbes can decrease the sorption of Ni^{2+} and Cd^{2+} to crushed volcanic tuff. Microbes increased the mobility of Ni^{2+} in the crushed tuff, but not to the extent predicted by the batch equilibration studies. The findings of decreased sorption and increased transport confirm studies by Chanmugathas and Bollag (1989), Burke et al. (1991), Gerringa (1990), and Wildung (1979). Our findings that microbes decrease dispersion are contrary to past studies showing that microbes increase dispersion as well as clogging pores (Taylor and Jaffé, 1990).

All of the observed effects may be of significance to prediction and management of cationic transition-metal fate and transport. For example, the rate of transport of metals from waste storage areas may be increased if microbial populations are present. Decreased dispersion caused by the presence of microbes may result in solutes occupying a smaller, more concentrated region.

The presence of microbes may also result in increased availability of metals to plants and other organisms in the soil. Although some metals in the solution may be used by organisms, increased concentrations of toxic metals may cause decreased growth and increased mortality. The ability of microbes to increase the availability of metals may be much greater than would be predicted by the results of this study, since the microbes may affect microenvironments surrounding the cells to a greater extent than they affect the bulk solution.

Although we did not study the phenomenon, we suspect that microbial effects on metal sorption may be more pronounced in nutrient-amended systems. Thus care should be taken that bioremediation of organic contaminants does not cause increased metal mobility.

Unlike Wildung (1979), who determined that Ni^{2+} transport was increased by microbial chelators, decreased sorption in our batch experiments was probably caused by a microbially influenced pH decrease. Thus, in stagnant systems or systems with a small water flux, as in many ground-water and vadose-zone systems, microbes that are present in the soils have the ability to change soil-water chemistry sufficiently to affect the transport of metals. The effect of microbes on metal retardation may be of less concern in systems with high fluid flux or low numbers of microbes because the natural buffering capacity of the soil may overcome the acid-producing capability of the microbes.

Our study has particular significance for waste sites located in volcanic tuff. We determined that the buffering capacity of volcanic tuff does not always exceed the acid-producing capability of the microbes and thus the microbially produced acid may affect the fate and transport of contaminants within the tuff. The microbes may also have had a greater affect on dispersion in the tuff than in other media that do not contain secondary porosity.

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Appendix A

Microbe Identification

APPENDIX A

MICROBE IDENTIFICATION

This appendix contains information on microbial identification including (1) the classification, (2) the identification number used in appendices B, D, and F, (3) the color, and (4) the gram stain type. The color and Gram Stain were described by Colette Kubichan in her laboratory notebook. The identification number is also used to identify cultures in the NMIMT microbiology laboratory (originally isolated by W. C. Ghiorse at Cornell University). The first part of the identification number refers to the depth in m from which the microbes were obtained. The following identifies each strain:

Microbe Number	Identity	Color	Gram Stain
50 T#2	*Pseudomonas*	Yellow	-
50 T#6	*Cryptococcus albidus* var. *albidus*	Pink	+
50 T#7	*Rhodococcus fascians* GC subgroup B	Pale yellow	+
50 T#8	*Pseudomonas*	Yellow	+
50 T#9	*Clavibacter michiganense insidiosum*	White	+
50 T#11	Unidentified yeast	White	+
50 T#12	*Pseudomonas*	Yellow	-
50 T#13	*Pseudomonas*	Yellow	-
150 T#3	*Arthrobacter oxydans*	White	-
175 T#1	*Arthrobacter oxydans*	White	-
175 T#2	*Arthrobacter oxydans*	White	-
175 T#3	*Bacillus megaterium*	White	+
175 T#6	*Arthrobacter oxydans*	White	-
175 T#7	*Candida famata*	Pink	+
175 T#8	*Candida famata*	Pink	+

In addition to the classification given above, information that Microbial ID, Inc., provided is summarized below:

1. 175 T#7 and 175 T#8 are the same species and probably the same strain.

2. All *Pseudomonas* strains belong to the same species; this species was not in the MIDI data base at the time of identification. 50 T#2, 50 T#8 are the same subspecies, 50 T#12 and 50 T#13 are the same subspecies. All four *Pseudomonas* are different strains.
3. All *Arthrobacter oxydans* (150 T#3, 175 T#1, 175 T#2, and 175 T#6) belong to the same species. 175 T#6 belongs to a different subspecies. All are different strains.
4. 50 T#11 may be a *Cryptococcus* or *Candida* strain.

Microbes designated 125 T#1, a designation not used in the original samples from Ghiorse, were used in some of the experiments. This designation may have resulted from mislabeling a culture of 175 T#2, since the fatty-acid profile was identical to 175 T#2.

Appendix B

Ni Batch Equilibrium Experiments

APPENDIX B

NICKEL BATCH EQUILIBRIUM EXPERIMENTS

This appendix consists of data for experiments performed to determine Ni^{2+} sorption to crushed tuff under a variety of conditions. Most experiments presented in this appendix contain both Ni^{2+} concentrations and optical densities of the microbial suspension used to inoculate the batch equilibrium samples. The concentrations of the standards are given under the heading 'initial concentration'. The standard concentrations were double the sample 'initial concentrations' since half of the solution in the samples consisted of the Ni^{2+} standard. The other half of the solution consisted of microbial suspension or sterile $CaCl_2$ solution. Experiments designated with an 'L' (L2 and L3Az7) were performed to determine the effect of freeze-dried *B. megaterium*. Experiments designated with 'Az' (L3Az7 and Az8) were performed to determine the NaN_3 effect. Experiments designated with an 'N' (N1 to N8) were performed to determine the effect of a variety of freshly washed microbes. The experiment designated 'B1' was used to show the effect of buffered system on Ni^{2+} sorption in the presence and absence of microbes.

The results from these experiments are summarized in the Results and Discussion section of Chapter 2 under the subheading 'Ni Sorption Experiments'. Results from L3Az7 were used for Figure 1. Experiments Az8 and L3Az7 were used to determine the average k_d for NaN_3. Experiments L2 and L3Az7 were used to determine k_d for freeze-dried *B. megaterium*: Experiments N1 and N7 were used to find isotherms for *Candida famata* (175 T#7), *Rhodococcus facsians* (50 T#7), and an *Arthrobacter oxydans* (175 T#1).

Experiments N2 to N5 were used to obtain data points for Figure 2; data from samples with initial Ni^{2+} concentrations of 10 mg L^{-1} were used. Experiments N2, N5, N7, N8, L2, and L3Az7 were used to determine the average k_d for *B. megaterium*; only samples with initial Ni^{2+} concentrations of 10 to 11 mg L^{-1} were used. Experiments N2 to N7, L2, L3, and B1 were used to find to find average k_d for all 14 strains and to find the k_d's for *Arthrobacter* and *Pseudomonas* strains. Experiments N8 and L2 contain pH data that were not reported in Chapter 2. In experiment N8, I also performed a test in which I determined that Ni^{2+} sorption to tuff in sterile samples is similar to sorption in samples containing supernatant used to wash microbes. These results indicate that the supernatant resulting from washing the microbes does not include chelators or microbially produced acid that changes the sorption. The sample used to run this test is designated as 'supn 175 T#3' in the table. Experiment B1 was used to produce Figure 3 in Chapter 2.

In addition to information reported in Chapter 2, experiment N1 contains information showing that results from filtered supernatant were the same as unfiltered supernatant. Also, for most experiments, initial solution liquid scintillation counts were taken from both the original Pyrex mixing flask (Standard) and from solutions which were shaken in centrifuge tubes for 24 hours (CT Standard). Results obtained using the two types of solution counts resulted in the same k_d to two significant figures.

Experiment: L3Az7
Experiment Dates: 2/8/93 to 2/10/93

Objective: Determine Ni isotherms for samples inoculated with freeze-dried B. megaterium (175 T#3), and sterile samples with and and without NaN3.

No optical density readings taken.

#	Sample Type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	sterile, azide	2784.9	2751.8	1.0	2.66E-02	1.93E+00
2	sterile, azide	2593.5	2560.4	1.0	2.47E-02	1.94E+00
3	sterile, azide	2630.5	2597.4	1.0	2.51E-02	1.94E+00
4	sterile, azide	3271.5	3238.4	4.0	1.20E-01	7.71E+00
5	sterile, azide	3010.7	2977.6	4.0	1.11E-01	7.73E+00
6	sterile, azide	2880.1	2847.0	4.0	1.06E-01	7.74E+00
7	sterile, azide	3835.9	3802.8	7.0	2.34E-01	1.34E+01
8	sterile, azide	3847.7	3814.6	7.0	2.34E-01	1.34E+01
9	sterile, azide	4267.2	4234.1	7.0	2.60E-01	1.34E+01
10	sterile, azide	2329.9	2296.8	10.0	3.67E-01	1.91E+01
11	sterile, azide	2332.6	2299.5	10.0	3.67E-01	1.91E+01
12	sterile, azide	2811.4	2778.3	10.0	4.44E-01	1.90E+01
13	Lyoph 175 T3	20719.2	20686.1	1.0	2.00E-01	1.59E+00
14	Lyoph 175 T3	20838.5	20805.4	1.0	2.01E-01	1.59E+00
15	Lyoph 175 T3	21193.7	21160.6	1.0	2.04E-01	1.58E+00
16	Lyoph 175 T3	23107.0	23073.9	4.0	8.57E-01	6.24E+00
17	Lyoph 175 T3	24924.5	24891.4	4.0	9.24E-01	6.10E+00
18	Lyoph 175 T3	22047.9	22014.8	4.0	8.17E-01	6.32E+00
19	Lyoph 175 T3	24048.4	24015.3	7.0	1.48E+00	1.10E+01
20	Lyoph 175 T3	23452.3	23419.2	7.0	1.44E+00	1.10E+01
21	Lyoph 175 T3	24346.1	24313.0	7.0	1.49E+00	1.09E+01
22	Lyoph 175 T3	13501.5	13468.4	10.0	2.15E+00	1.56E+01
23	Lyoph 175 T3	14139.3	14106.2	10.0	2.25E+00	1.54E+01
24	Lyoph 175 T3	14599.6	14566.5	10.0	2.33E+00	1.52E+01
25	sterile, nonazid	851.2	818.1	1.0	7.90E-03	1.97E+00
26	sterile, nonazid	744.0	710.9	1.0	6.87E-03	1.97E+00
27	sterile, nonazid	704.8	671.7	1.0	6.49E-03	1.98E+00
28	sterile, nonazid	797.5	764.4	4.0	2.84E-02	7.90E+00
29	sterile, nonazid	893.1	860.0	4.0	3.19E-02	7.89E+00
30	sterile, nonazid	720.7	687.6	4.0	2.55E-02	7.90E+00
31	sterile, nonazid	864.2	831.1	7.0	5.11E-02	1.38E+01
32	sterile, nonazid	788.2	755.1	7.0	4.64E-02	1.38E+01
33	sterile, nonazid	784.8	751.7	7.0	4.62E-02	1.38E+01
34	sterile, nonazid	405.9	372.8	10.0	5.96E-02	1.98E+01
35	sterile, nonazid	590.2	557.1	10.0	8.90E-02	1.97E+01
36	sterile, nonazid	499.0	465.9	10.0	7.44E-02	1.97E+01
37	Standard	205260.0	205226.9	2.0		
38	Standard	207904.0	207870.9	2.0		
39	Standard	207950.0	207916.9	2.0		
40	Standard	214016.0	213982.9	8.0		
41	Standard	215625.0	215591.9	8.0		

Experiment: L3Az7
Experiment Dates: 2/8/93 to 2/10/93

Objective: Determine Ni isotherms for samples inoculated with freeze-dried B. megaterium (175 T#3), and sterile samples with and and without NaN3.

No optical density readings taken.

#	Sample Type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
42	Standard	216991.0	216957.9	8.0		
43	Standard	225937.0	225903.9	14.0		
44	Standard	228946.0	228912.9	14.0		
45	Standard	228826.0	228792.9	14.0		
46	Standard	124182.0	124148.9	20.0		
47	Standard	125405.0	125371.9	20.0		
48	Standard	126072.0	126038.9	20.0		

Experiment: L2
Experiment Dates: 9/16/92 to 9/18/92

Objective: Determine Ni isotherms for samples inoculated with freeze-dried 175 T#3 We also have plate counts and direct counts of this experiment.

Optical Density of Lyophilized 175 T#3: 0.78

#	Sample Type	CPM/mL	CPM/mL - Background	pH	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	sterile	259.0	225.9	8.45	1.0	1.19E-02	1.89E+00
2	sterile	222.6	189.5		1.0	9.99E-03	1.90E+00
3	sterile	243.3	210.2		1.0	1.11E-02	1.89E+00
4	sterile	238.0	204.9	8.52	4.0	4.25E-02	7.85E+00
5	sterile	272.4	239.3		4.0	4.97E-02	7.84E+00
6	sterile	266.0	232.9		4.0	4.83E-02	7.84E+00
7	sterile	231.8	198.7	8.55	7.0	8.08E-02	1.46E+01
8	sterile	231.6	198.5		7.0	8.08E-02	1.46E+01
9	sterile	227.3	194.2		7.0	7.90E-02	1.46E+01
10	sterile	219.6	186.5	8.52	10.0	1.03E-01	1.97E+01
11	sterile	214.0	180.9		10.0	1.00E-01	1.97E+01
12	sterile	231.7	198.6		10.0	1.10E-01	1.96E+01
13	Lyoph175 T#3	3861.8	3828.7	7.26	1.0	2.02E-01	1.51E+00
14	Lyoph175 T#3	3597.9	3564.8		1.0	1.88E-01	1.54E+00
15	Lyoph175 T#3	3589.6	3556.5		1.0	1.87E-01	1.54E+00
16	Lyoph175 T#3	3834.3	3801.2	7.10	4.0	7.89E-01	6.36E+00
17	Lyoph175 T#3	3983.2	3950.1		4.0	8.20E-01	6.30E+00
18	Lyoph175 T#3	5649.0	5615.9		4.0	1.17E+00	5.60E+00
19	Lyoph175 T#3	3019.4	2986.3	7.45	7.0	1.21E+00	1.24E+01
20	Lyoph175 T#3	4729.6	4696.5		7.0	1.91E+00	1.10E+01
21	Lyoph175 T#3	3625.9	4696.5		7.0	1.91E+00	1.10E+01
22	Lyoph175 T#3	2977.8	2944.7	7.45	10.0	1.63E+00	1.66E+01
23	Lyoph175 T#3	2778.5	2745.4		10.0	1.52E+00	1.68E+01
24	Lyoph175 T#3	3634.4	3601.3		10.0	2.00E+00	1.59E+01
25	CT Standard	35911.9	35878.8		2.0		
26	CT Standard	36863.8	36830.7		2.0		
27	CT Standard	38433.0	38399.9		8.0		
28	CT Standard	38103.2	38070.1		8.0		
29	CT Standard	36282.8	36249.7		14.0		
30	CT Standard	36556.2	36523.1		14.0		
31	CT Standard	35720.2	35687.1		20.0		
32	CT Standard	35941.9	35908.8		20.0		
33	Standard	37982.5	37949.4		2.0		
34	Standard	37989.2	37956.1		2.0		
35	Standard	38451.2	38418.1		8.0		
36	Standard	38705.2	38672.1		8.0		
37	Standard	34523.3	34490.2		14.0		
38	Standard	34368.6	34335.5		14.0		
39	Standard	36093.6	36060.5		20.0		
40	Standard	36078.0	36044.9		20.0		

Experiment: Az8
Experiment Dates: 2/17/93 to 2/19/93

Objective: Determine Ni isotherms for samples containing NaN3.

Optical Density: N/A

#	Sample Type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	sterile, azide	2436.4	2401.1	1.02	2.32E-02	1.98E+00
2	sterile, azide	2870.4	2835.1	1.02	2.74E-02	1.97E+00
3	sterile, azide	2975.6	2940.3	1.02	2.84E-02	1.97E+00
4	sterile, azide	3186.5	3151.2	3.98	1.17E-01	7.68E+00
5	sterile, azide	3007.9	2972.6	3.98	1.10E-01	7.70E+00
6	sterile, azide	3311.8	3276.5	3.98	1.22E-01	7.67E+00
7	sterile, azide	4281.6	4246.3	7.05	2.61E-01	1.35E+01
8	sterile, azide	4393.2	4357.9	7.05	2.68E-01	1.35E+01
9	sterile, azide	4145.6	4110.3	7.05	2.53E-01	1.35E+01
10	sterile, azide	2674.1	2638.8	9.97	4.22E-01	1.90E+01
11	sterile, azide	2609.2	2573.9	9.97	4.11E-01	1.90E+01
12	sterile, azide	2639.3	2604.0	9.97	4.16E-01	1.90E+01
13	sterile, no azide	902.8	867.5	1.02	8.38E-03	2.01E+00
14	sterile, no azide	794.4	759.1	1.02	7.33E-03	2.01E+00
15	sterile, no azide	774.8	739.5	1.02	7.14E-03	2.02E+00
16	sterile, no azide	812.9	777.6	3.98	2.89E-02	7.86E+00
17	sterile, no azide	876.0	840.7	3.98	3.12E-02	7.86E+00
18	sterile, no azide	809.8	774.5	3.98	2.87E-02	7.86E+00
19	sterile, no azide	820.7	785.4	7.05	4.83E-02	1.39E+01
20	sterile, no azide	943.3	908.0	7.05	5.58E-02	1.39E+01
21	sterile, no azide	979.2	943.9	7.05	5.80E-02	1.39E+01
22	sterile, no azide	605.7	570.4	9.97	9.11E-02	1.96E+01
23	sterile, no azide	636.9	601.6	9.97	9.61E-02	1.96E+01
24	sterile, no azide	624.3	589.0	9.97	9.41E-02	1.96E+01
25	Standard	210030.0	209994.7	2.04		
26	Standard	212305.0	212269.7	2.04		
27	Standard	211758.0	211722.7	2.04		
28	Standard	214386.0	214350.7	7.97		
29	Standard	214148.0	214112.7	7.97		
30	Standard	215353.0	215317.7	7.97		
31	Standard	229202.0	229166.7	14.11		
32	Standard	228386.0	228350.7	14.11		
33	Standard	231379.0	231343.7	14.11		
34	Standard	124521.0	124485.7	19.95		
35	Standard	124404.0	124368.7	19.95		
36	Standard	125751.0	125715.7	19.95		

Batch Experiment: N1
Experiment Dates: 10/23/91 to 10/25/91

Objective: Determine Ni isotherms for samples inoculated with 50 T#7 and 175 T#1.

Optical Densities:
50 T#7: 0.80
175 T#1: 0.80

#	Sample Type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	CT Standard	35903.0	35869.9	0.04		
2	CT Standard	35530.8	35497.7	0.04		
3	CT Standard	34937.9	34904.8	0.04		
4	CT Standard	37216.3	37183.2	0.4		
5	CT Standard	36595.5	36562.4	0.4		
6	CT Standard	35559.5	35526.4	0.4		
7	CT Standard	35159.9	35126.8	4.0		
8	CT Standard	35695.7	35662.6	4.0		
9	CT Standard	35402.8	35369.7	4.0		
10	CT Standard	35723.7	35690.6	40.0		
11	CT Standard	36096.6	36063.5	40.0		
12	CT Standard	35858.3	35825.2	40.0		
13	CT Standard	37499.4	37466.3	400.0		
14	CT Standard	37149.1	37116.0	400.0		
15	CT Standard	37179.2	37146.1	400.0		
16	50 T#7	774.7	741.6	0.02	8.37E-04	3.83E-02
17	50 T#7	744.2	711.1	0.02	8.03E-04	3.84E-02
18	50 T#7	716.4	683.3	0.02	7.72E-04	3.85E-02
19	50 T#7	768.8	735.7	0.2	8.08E-03	3.84E-01
20	50 T#7	692.6	659.5	0.2	7.24E-03	3.86E-01
21	50 T#7	610.2	577.1	0.2	6.34E-03	3.87E-01
22	50 T#7	573.2	540.1	2.0	6.11E-02	3.88E+00
23	50 T#7	590.7	557.6	2.0	6.30E-02	3.87E+00
24	50 T#7	621.4	588.3	2.0	6.65E-02	3.87E+00
25	50 T#7	1171.8	1138.7	20.0	1.27E+00	3.75E+01
26	50 T#7	1164.4	1131.3	20.0	1.26E+00	3.75E+01
27	50 T#7	1138.5	1105.4	20.0	1.23E+00	3.75E+01
28	50 T#7	2759.9	2726.8	200.0	2.93E+01	3.41E+02
29	50 T#7	2838.3	2805.2	200.0	3.01E+01	3.40E+02
30	50 T#7	2861.0	2827.9	200.0	3.04E+01	3.39E+02
31	175 T#1	1167.3	1134.2	0.02	1.28E-03	3.74E-02
32	175 T#1	1067.5	1034.4	0.02	1.17E-03	3.77E-02
33	175 T#1	1031.9	998.8	0.02	1.13E-03	3.77E-02
34	175 T#1	1152.4	1119.3	0.2	1.23E-02	3.75E-01
35	175 T#1	1197.2	1164.1	0.2	1.28E-02	3.74E-01
36	175 T#1	1207.6	1174.5	0.2	1.29E-02	3.74E-01
37	175 T#1	1075.7	1042.6	2.0	1.18E-01	3.76E+00
38	175 T#1	949.5	916.4	2.0	1.04E-01	3.79E+00
39	175 T#1	1089.5	1056.4	2.0	1.19E-01	3.76E+00
40	175 T#1	1601.7	1568.6	20.0	1.75E+00	3.65E+01
41	175 T#1	1648.3	1615.2	20.0	1.80E+00	3.64E+01
42	175 T#1	1708.6	1675.5	20.0	1.87E+00	3.63E+01
43	175 T#1	3254.9	3221.8	200.0	3.46E+01	3.31E+02

Batch Experiment: N1
Experiment Dates: 10/23/91 to 10/25/91

Objective: Determine Ni isotherms for samples inoculated with 50 T#7 and 175 T#1.

Optical Densities:		
	50 T#7:	0.80
	175 T#1:	0.80

#	Sample Type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
44	175 T#1	3127.4	3094.3	200.0	3.32E+01	3.34E+02
45	175 T#1	3062.4	3029.3	200.0	3.25E+01	3.35E+02
46	sterile	232.1	199.0	0.02	2.25E-04	3.96E-02
47	sterile	272.3	239.2	0.02	2.70E-04	3.95E-02
48	sterile	266.2	233.1	0.02	2.63E-04	3.95E-02
49	sterile	265.0	231.9	0.2	2.55E-03	3.95E-01
50	sterile	249.0	215.9	0.2	2.37E-03	3.95E-01
51	sterile	260.2	227.1	0.2	2.49E-03	3.95E-01
52	sterile	221.1	188.0	2.0	2.13E-02	3.96E+00
53	sterile	224.6	191.5	2.0	2.17E-02	3.96E+00
54	sterile	236.0	202.9	2.0	2.29E-02	3.95E+00
55	sterile	307.7	274.6	20.0	3.06E-01	3.94E+01
56	sterile	294.4	261.3	20.0	2.92E-01	3.94E+01
57	sterile	280.4	247.3	20.0	2.76E-01	3.94E+01
58	sterile	1495.2	1462.1	200.0	1.57E+01	3.69E+02
59	sterile	1685.5	1652.4	200.0	1.77E+01	3.65E+02
60	sterile	1728.5	1695.4	200.0	1.82E+01	3.64E+02
61	bkgd w/ tuff	36.0	2.9			
62	bkgd w/ tuff	34.1	1.0			
63	bkgd w/ tuff	30.3	-2.8			
64	Standard	36227.0	36193.9	0.04		
65	Standard	36349.6	36316.5	0.04		
66	Standard	36518.0	36484.9	0.04		
67	Standard	37698.1	37665.0	0.4		
68	Standard	37845.7	37812.6	0.4		
69	Standard	37826.9	37793.8	0.4		
70	Standard	35705.6	35672.5	4.0		
71	Standard	35643.1	35610.0	4.0		
72	Standard	35472.8	35439.7	4.0		
73	Standard	35760.4	35727.3	40.0		
74	Standard	35836.7	35803.6	40.0		
75	Standard	35735.3	35702.2	40.0		
76	Standard	36885.0	36851.9	400.0		
77	Standard	36868.0	36834.9	400.0		
78	Standard	36767.5	36734.4	400.0		
79	filtered 16	808.0	774.9			
80	filtered 17	762.1	729.0			
81	filtered 18	744.2	711.1			
82	filtered 28	2716.9	2683.8			
83	filtered 29	2897.2	2864.1			
84	filtered 30	2987.4	2954.3			
85	filtered 31	1195.0	1161.9			
86	filtered 32	1076.2	1043.1			

Batch Experiment: N1
Experiment Dates: 10/23/91 to 10/25/91

Objective: Determine Ni isotherms for samples inoculated with 50 T#7 and 175 T#1.

Optical Densities: 50 T#7: 0.80
175 T#1: 0.80

#	Sample Type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
87	filtered 33	1056.3	1023.2			
88	air/no Sc. C.	28.4				
89	bkgr	34.3				
90	bkgr	30.5				
91	bkgr	33.2				

Batch Experiment: N2
Experiment Dates: 11/20/91 to 11/22/91

Objective: Find the effects of 50 T#2, 50 T#6, 50 T#7, 50 T#9, 50 T#12, 50 T#13, 175 T#3, and 175 T#7 on Ni sorption to crushed tuff.

Optical Densities:
50 T#2: 0.56
50 T#6: 0.46
50 T#7: 1.30
50 T#9: 1.00
50 T#12: 0.80
50 T#13: 0.80
175 T#3: 0.75
175 T#7: 0.75

#	Sample Type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	sterile	339.0	305.9	10.00	1.82E-01	1.96E+01
2	sterile	313.3	280.2	10.00	1.67E-01	1.97E+01
3	sterile	315.5	282.4	10.00	1.68E-01	1.97E+01
4	50 T#2	704.3	671.2	10.00	4.00E-01	1.92E+01
5	50 T#2	688.9	655.8	10.00	3.91E-01	1.92E+01
6	50 T#2	717.3	684.2	10.00	4.08E-01	1.92E+01
7	50 T#6	925.3	892.2	10.00	5.31E-01	1.89E+01
8	50 T#6	850.0	816.9	10.00	4.87E-01	1.90E+01
9	50 T#6	820.8	787.7	10.00	4.69E-01	1.91E+01
10	50 T#7	777.4	744.3	10.00	4.43E-01	1.91E+01
11	50 T#7	588.4	555.3	10.00	3.31E-01	1.93E+01
12	50 T#7	680.2	647.1	10.00	3.85E-01	1.92E+01
13	50 T#9	603.9	570.8	10.00	3.40E-01	1.93E+01
14	50 T#9	552.8	519.7	10.00	3.10E-01	1.94E+01
15	50 T#9	717.4	684.3	10.00	4.08E-01	1.92E+01
16	50 T#12	577.6	544.5	10.00	3.24E-01	1.94E+01
17	50 T#12	617.2	584.1	10.00	3.48E-01	1.93E+01
18	50 T#12	563.6	530.5	10.00	3.16E-01	1.94E+01
19	50 T#13	706.6	673.5	10.00	4.01E-01	1.92E+01
20	50 T#13	697.3	664.2	10.00	3.96E-01	1.92E+01
21	50 T#13	726.8	693.7	10.00	4.13E-01	1.92E+01
22	175 T#3	1176.0	1142.9	10.00	6.81E-01	1.86E+01
23	175 T#3	1196.1	1163.0	10.00	6.93E-01	1.86E+01
24	175 T#3	1125.4	1092.3	10.00	6.51E-01	1.87E+01
25	175 T#7	1113.0	1079.9	10.00	6.43E-01	1.87E+01
26	175 T#7	1100.3	1067.2	10.00	6.36E-01	1.87E+01
27	175 T#7	1077.5	1044.4	10.00	6.22E-01	1.88E+01
28	CT Standard	33814.1	33781.0	20.00		
29	CT Standard	33571.3	33538.2	20.00		
30	CT Standard	33444.6	33411.5	20.00		
31	Standard	33427.7	33394.6	20.00		
32	Standard	33589.6	33556.5	20.00		
33	Standard	33519.0	33485.9	20.00		

Batch Experiment: N3
Experiment Dates: 12/2/91 to 12/4/91

Objective: Find the effects of 50 T#8, 150 T#3, 175 T#2, and 175 T#8 on Ni sorption to crushed tuff.

Optical Densities:				
	50 T#8:	0.85	175 T#2:	0.85
	150 T#3:	0.75	175 T#8:	0.80

#	Sample Type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	sterile	248.4	215.3	10.00	1.29E-01	1.97E+01
2	sterile	259.9	226.8	10.00	1.36E-01	1.97E+01
3	sterile	309.1	276.0	10.00	1.66E-01	1.97E+01
4	50 T#8	506.9	473.8	10.00	2.85E-01	1.94E+01
5	50 T#8	610.9	577.8	10.00	3.47E-01	1.93E+01
6	50 T#8	667.4	634.3	10.00	3.81E-01	1.92E+01
7	150 T#3	366.8	333.7	10.00	2.01E-01	1.96E+01
8	150 T#3	364.0	330.9	10.00	1.99E-01	1.96E+01
9	150 T#3	370.5	337.4	10.00	2.03E-01	1.96E+01
10	175 T#2	532.4	499.3	10.00	3.00E-01	1.94E+01
11	175 T#2	481.5	448.4	10.00	2.70E-01	1.95E+01
12	175 T#2	545.1	512.0	10.00	3.08E-01	1.94E+01
13	175 T#8	768.2	735.1	10.00	4.42E-01	1.91E+01
14	175 T#8	721.7	688.6	10.00	4.14E-01	1.92E+01
15	175 T#8	779.6	746.5	10.00	4.49E-01	1.91E+01
16	CT Standard	32918.9	32885.8	20.00		
17	CT Standard	33997.1	33964.0	20.00		
18	CT Standard	32950.9	32917.8	20.00		
19	Standard	33350.8	33317.7	20.00		
20	Standard	33352.9	33319.8	20.00		
21	Standard	33264.8	33231.7	20.00		

Batch Experiment: N4
Experiment Dates: 12/12/91 to 12/14/91

Objective: Find the effect of 175 T#1 and 175 T#6 on Ni sorption to crushed tuff.

Optical Densities: 175 T#1: 0.80
175 T#6: 0.75

#	Sample Type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	sterile	371.1	338.0	10.00	1.92E-01	1.96E+01
2	sterile	379.6	346.5	10.00	1.97E-01	1.96E+01
3	sterile	377.8	344.7	10.00	1.96E-01	1.96E+01
4	175 T#1	1556.3	1523.2	10.00	8.66E-01	1.83E+01
5	175 T#1	1829.0	1795.9	10.00	1.02E+00	1.80E+01
6	175 T#1	1277.4	1244.3	10.00	7.08E-01	1.86E+01
7	175 T#6	1405.0	1371.9	10.00	7.80E-01	1.84E+01
8	175 T#6	1448.1	1415.0	10.00	8.05E-01	1.84E+01
9	175 T#6	1442.2	1409.1	10.00	8.01E-01	1.84E+01
10	CT Standard	35518.8	35485.7	20.00		
11	CT Standard	33765.8	33732.7	20.00		
12	CT Standard	36301.6	36268.5	20.00		
13	Standard	32222.6	32189.5	20.00		
14	Standard	32138.1	32105.0	20.00		
15	Standard	30969.2	30936.1	20.00		

Batch Experiment: N5
Experiment Dates: 12/22/91 to 12/24/91

Objective: Find the effect of 125 T#1 and 175 T#1, 175 T#3, and 175 T#6 on Ni sorption to crushed tuff.

Optical Densities:				
	125 T#1:	0.80	175 T#3:	0.75
	175 T#1:	0.85	175 T#6:	0.80

#	Sample Type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	sterile	294.4	261.3	10.00	1.42E-01	1.97E+01
2	sterile	188.2	155.1	10.00	8.40E-02	1.98E+01
3	sterile	220.7	187.6	10.00	1.02E-01	1.98E+01
4	125 T#1	1100.8	1067.7	10.00	5.78E-01	1.88E+01
5	125 T#1	1039.5	1006.4	10.00	5.45E-01	1.89E+01
6	125 T#1	1209.3	1176.2	10.00	6.37E-01	1.87E+01
7	175 T#1	1371.2	1338.1	10.00	7.25E-01	1.86E+01
8	175 T#1	1278.4	1245.3	10.00	6.75E-01	1.87E+01
9	175 T#1	1367.1	1334.0	10.00	7.23E-01	1.86E+01
10	175 T#3	4053.3	4020.2	10.00	2.18E+00	1.56E+01
11	175 T#3	4047.5	4014.4	10.00	2.17E+00	1.57E+01
12	175 T#3	3868.8	3835.7	10.00	2.08E+00	1.58E+01
13	175 T#6	2136.0	2102.9	10.00	1.14E+00	1.77E+01
14	175 T#6	2918.4	2885.3	10.00	1.56E+00	1.69E+01
15	175 T#6	2407.3	2374.2	10.00	1.29E+00	1.74E+01
16	CT Standard	38153.5	38120.4	20.00		
17	CT Standard	36587.3	36554.2	20.00		
18	CT Standard	36105.6	36072.5	20.00		
19	Standard	37275.5	37242.4	20.00		
20	Standard	36759.3	36726.2	20.00		
21	Standard	36142.2	36109.1	20.00		

Batch Experiment: N6
Experiment Dates: 12/27/91 to 12/29/91

Objective: Find the effect of 50 T#11 and 175 T#7 on Ni sorption to crushed tuff.

Optical Densities: 50 T#11: 0.36
175 T#7: 0.80

#	Sample Type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	sterile	290.4	257.3	11.25	1.64E-01	2.22E+01
2	sterile	301.1	268.0	11.25	1.71E-01	2.22E+01
3	sterile	288.6	255.5	11.25	1.63E-01	2.22E+01
4	50 T#11	1490.4	1457.3	11.25	9.32E-01	2.06E+01
5	50 T#11	1235.7	1202.6	11.25	7.69E-01	2.10E+01
6	50 T#11	1119.1	1086.0	11.25	6.94E-01	2.11E+01
7	175 T#7	2200.5	2167.4	11.25	1.39E+00	1.97E+01
8	175 T#7	3125.8	3092.7	11.25	1.98E+00	1.85E+01
9	175 T#7	2115.0	2081.9	11.25	1.33E+00	1.98E+01
10	CT Standard	34736.0	34702.9	22.50		
11	CT Standard	35747.5	35714.4	22.50		
12	CT Standard	35195.0	35161.9	22.50		
13	Standard	35303.0	35269.9	22.50		
14	Standard	35421.5	35388.4	22.50		
15	Standard	35901.3	35868.2	22.50		

Batch Experiment: N7
Experiment Dates: 1/13/92 to 1/15/92

Objective: Determine Ni isotherms for samples inoculated with175 T#3 and 175 T#7.

Optical Densities: 175 T#3: 0.80
175 T#7: 0.80

#	Sample type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	sterile	254.0	220.9	0.01	1.18E-04	2.23E-02
2	sterile	278.0	244.9	0.01	1.31E-04	2.22E-02
3	sterile	300.0	266.9	0.01	1.42E-04	2.22E-02
4	sterile	286.0	252.9	0.11	1.32E-03	2.22E-01
5	sterile	286.0	252.9	0.11	1.32E-03	2.22E-01
6	sterile	285.0	251.9	0.11	1.31E-03	2.22E-01
7	sterile	272.0	238.9	1.13	1.32E-02	2.22E+00
8	sterile	292.0	258.9	1.13	1.43E-02	2.22E+00
9	sterile	283.0	249.9	1.13	1.38E-02	2.22E+00
10	sterile	319.0	285.9	11.25	1.55E-01	2.22E+01
11	sterile	362.0	328.9	11.25	1.78E-01	2.21E+01
12	sterile	437.0	403.9	11.25	2.19E-01	2.21E+01
13	sterile	971.0	937.9	112.50	4.61E+00	2.16E+02
14	sterile	1064.0	1030.9	112.50	5.07E+00	2.15E+02
15	sterile	848.0	814.9	112.50	4.01E+00	2.17E+02
16	175 T#3	3870.0	3836.9	0.01	2.05E-03	1.84E-02
17	175 T#3	4116.0	4082.9	0.01	2.18E-03	1.81E-02
18	175 T#3	4291.0	4257.9	0.01	2.27E-03	1.80E-02
19	175 T#3	3864.0	3830.9	0.11	2.00E-02	1.85E-01
20	175 T#3	4053.0	4019.9	0.11	2.09E-02	1.83E-01
21	175 T#3	4130.0	4096.9	0.11	2.13E-02	1.82E-01
22	175 T#3	4475.0	4441.9	1.13	2.45E-01	1.76E+00
23	175 T#3	4615.0	4581.9	1.13	2.53E-01	1.74E+00
24	175 T#3	4391.0	4357.9	1.13	2.40E-01	1.77E+00
25	175 T#3	4382.0	4348.9	11.25	2.35E+00	1.78E+01
26	175 T#3	3911.0	3877.9	11.25	2.10E+00	1.83E+01
27	175 T#3	4103.0	4069.9	11.25	2.20E+00	1.81E+01
28	175 T#3	6927.0	6893.9	112.50	3.39E+01	1.57E+02
29	175 T#3	7086.0	7052.9	112.50	3.47E+01	1.56E+02
30	175 T#3	6708.0	6674.9	112.50	3.28E+01	1.59E+02
31	175 T#7	1962.0	1928.9	0.01	1.03E-03	2.04E-02
32	175 T#7	1957.0	1923.9	0.01	1.03E-03	2.04E-02
33	175 T#7	2028.0	1994.9	0.01	1.06E-03	2.04E-02
34	175 T#7	2237.0	2203.9	0.11	1.15E-02	2.02E-01
35	175 T#7	2239.0	2205.9	0.11	1.15E-02	2.02E-01
36	175 T#7	2263.0	2229.9	0.11	1.16E-02	2.02E-01
37	175 T#7	1992.0	1958.9	1.13	1.08E-01	2.03E+00
38	175 T#7	2041.0	2007.9	1.13	1.11E-01	2.03E+00
39	175 T#7	2102.0	2068.9	1.13	1.14E-01	2.02E+00
40	175 T#7	2251.0	2217.9	11.25	1.20E+00	2.01E+01
41	175 T#7	2310.0	2276.9	11.25	1.23E+00	2.00E+01
42	175 T#7	2632.0	2598.9	11.25	1.41E+00	1.97E+01
43	175 T#7	5426.0	5392.9	112.50	2.65E+01	1.72E+02

Batch Experiment: N7
Experiment Dates: 1/13/92 to 1/15/92

Objective: Determine Ni isotherms for samples inoculated with175 T#3 and 175 T#7.

Optical Densities: 175 T#3: 0.80
175 T#7: 0.80

#	Sample type	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
44	175 T#7	4858.0	4824.9	112.50	2.37E+01	1.78E+02
45	175 T#7	6463.0	6429.9	112.50	3.16E+01	1.62E+02
46	CT Standard	42130.0	42096.9	0.02		
47	CT Standard	42133.0	42099.9	0.02		
48	CT Standard	42358.0	42324.9	0.02		
49	CT Standard	43747.0	43713.9	0.23		
50	CT Standard	42715.0	42681.9	0.23		
51	CT Standard	43207.9	43174.8	0.23		
52	CT Standard	40828.0	40794.9	2.25		
53	CT Standard	40911.0	40877.9	2.25		
54	CT Standard	40698.0	40664.9	2.25		
55	CT Standard	41876.0	41842.9	22.50		
56	CT Standard	41209.0	41175.9	22.50		
57	CT Standard	41673.0	41639.9	22.50		
58	CT Standard	45701.0	45667.9	225.00		
59	CT Standard	46101.0	46067.9	225.00		
60	CT Standard	45577.0	45543.9	225.00		
61	Standard	42614.0	42580.9	0.02		
62	Standard	42798.0	42764.9	0.02		
63	Standard	41444.0	41410.9	0.02		
64	Standard	43179.0	43145.9	0.23		
65	Standard	13306.0	13272.9	0.23		
66	Standard	43287.0	43253.9	0.23		
67	Standard	41536.0	41502.9	2.25		
68	Standard	41103.0	41069.9	2.25		
69	Standard	40857.0	40823.9	2.25		
70	Standard	41780.0	41746.9	22.50		
71	Standard	41755.0	41721.9	22.50		
72	Standard	41702.0	41668.9	22.50		
73	Standard	46006.9	45973.8	225.00		
74	Standard	46732.0	46698.9	225.00		
75	Standard	46181.0	46147.9	225.00		

Batch Experiment: N8
Experiment Dates: 2/3/92 to 2/5/92

Objective: Find the effect of 175 T#3 and 175 T#1, 175 T#6, and 175 T#7 on Ni Sorption to crushed tuff.

Optical Densities:				
	175 T#3:	0.80	175 T#6	0.72
	175 T#1:	0.81	175 T#7	0.24

#	Sample Type	pH	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	sterile	7.69	240.0	215.0	0.02	2.88E-04	4.47E-02
2	sterile	7.71	225.0	200.0	0.02	2.68E-04	4.48E-02
3	sterile	7.69	214.0	189.0	0.02	2.54E-04	4.48E-02
4	sterile	7.68	234.0	209.0	0.23	2.48E-03	4.48E-01
5	sterile	7.82	235.0	210.0	0.23	2.49E-03	4.48E-01
6	sterile	7.67	233.0	208.0	0.23	2.47E-03	4.48E-01
7	sterile	7.66	248.0	223.0	2.27	2.61E-02	4.48E+00
8	sterile	7.63	223.0	198.0	2.27	2.32E-02	4.48E+00
9	sterile	7.80	237.0	212.0	2.27	2.48E-02	4.48E+00
10	sterile	7.84	329.0	304.0	22.65	3.05E-01	4.47E+01
11	sterile	7.72	366.0	341.0	22.65	3.43E-01	4.46E+01
12	sterile	7.72	365.0	340.0	22.65	3.42E-01	4.46E+01
13	sterile	7.07	2906.0	2881.0	226.50	3.54E+01	3.82E+02
14	sterile	7.02	3095.0	3070.0	226.50	3.77E+01	3.78E+02
15	sterile	6.97	3326.0	3301.0	226.50	4.06E+01	3.72E+02
16	175 T#3	7.05	3085.0	3060.0	0.02	4.10E-03	3.71E-02
17	175 T#3	6.79	3056.0	3031.0	0.02	4.07E-03	3.72E-02
18	175 T#3	6.83	3396.0	3371.0	0.02	4.52E-03	3.63E-02
19	175 T#3	6.79	3949.0	3924.0	0.23	4.66E-02	3.60E-01
20	175 T#3	6.80	3994.0	3969.0	0.23	4.71E-02	3.59E-01
21	175 T#3	6.79	4134.0	4109.0	0.23	4.88E-02	3.55E-01
22	175 T#3	6.87	3717.0	3692.0	2.27	4.32E-01	3.67E+00
23	175 T#3	6.86	3771.0	3746.0	2.27	4.38E-01	3.65E+00
24	175 T#3	6.85	3526.0	3501.0	2.27	4.10E-01	3.71E+00
25	175 T#3	6.85	3502.0	3477.0	22.65	3.49E+00	3.83E+01
26	175 T#3	6.87	3403.0	3378.0	22.65	3.39E+00	3.85E+01
27	175 T#3	6.92	3698.0	3673.0	22.65	3.69E+00	3.79E+01
28	175 T#3	6.55	5946.0	5921.0	226.50	7.27E+01	3.08E+02
29	175 T#3	6.58	6744.0	6719.0	226.50	8.26E+01	2.88E+02
30	175 T#3	6.58	6552.0	6527.0	226.50	8.02E+01	2.93E+02
31	175 T#1	7.26	1947.0	1922.0	22.65	1.93E+00	4.14E+01
32	175 T#1	7.33	988.0	963.0	22.65	9.67E-01	4.34E+01
33	175 T#1	7.25	1134.0	1109.0	22.65	1.11E+00	4.31E+01
34	175 T#6	7.43	1045.0	1020.0	22.65	1.02E+00	4.33E+01
35	175 T#6	7.34	1221.0	1196.0	22.65	1.20E+00	4.29E+01
36	175 T#6	7.44	1346.0	1321.0	22.65	1.33E+00	4.26E+01
37	175 T#7	7.53	901.0	876.0	22.65	8.80E-01	4.35E+01
38	175 T#7	7.40	1154.0	1129.0	22.65	1.13E+00	4.30E+01
39	175 T#7	Not sampled		-	-	-	-
40	supn. 175 T#3	7.54	424.0	399.0	22.65	4.01E-01	4.45E+01
41	supn. 175 T#3	7.62	373.0	348.0	22.65	3.50E-01	4.46E+01

Batch Experiment: N8
Experiment Dates: 2/3/92 to 2/5/92

Objective: Find the effect of 175 T#3 and 175 T#1, 175 T#6, and 175 T#7 on Ni Sorption to crushed tuff.

Optical Densities: 175 T#3: 0.80 175 T#6 0.72
175 T#1: 0.81 175 T#7 0.24

#	Sample Type	pH	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
42	supn. 175 T#3	7.60	360.0	335.0	22.65	3.36E-01	4.46E+01
43	supn. 175 T#3	7.62	271.0	246.0	22.65	2.47E-01	4.48E+01
44	supn. 175 T#3	7.60	274.0	249.0	22.65	2.50E-01	4.48E+01
45	supn. 175 T#3	7.60	217.0	192.0	22.65	1.93E-01	4.49E+01
46	CT Standard		33626.0	33601.0	0.05		
47	CT Standard		33751.0	33726.0	0.05		
48	CT Standard		34008.0	33983.0	0.05		
49	CT Standard		37859.0	37834.0	0.45		
50	CT Standard		38368.0	38343.0	0.45		
51	CT Standard		38364.0	38339.0	0.45		
52	CT Standard		38576.0	38551.0	4.53		
53	CT Standard		38603.0	38578.0	4.53		
54	CT Standard		39018.0	38993.0	4.53		
55	CT Standard		45651.0	45626.0	45.30		
56	CT Standard		44595.0	44570.0	45.30		
57	CT Standard		45131.9	45106.9	45.30		
58	CT Standard		37202.0	37177.0	453.00		
59	CT Standard		36682.0	36657.0	453.00		
60	CT Standard		36798.0	36773.0	453.00		
61	Standard		34343.0	34318.0	0.05		
62	Standard		34138.0	34113.0	0.05		
63	Standard		34089.0	34064.0	0.05		
64	Standard		38589.0	38564.0	0.45		
65	Standard		38559.0	38534.0	0.45		
66	Standard		38377.0	38352.0	0.45		
67	Standard		39215.0	39190.0	4.53		
68	Standard		39192.0	39167.0	4.53		
69	Standard		38949.0	38924.0	4.53		
70	Standard		46113.0	46088.0	45.30		
71	Standard		46415.0	46390.0	45.30		
72	Standard		46704.0	46679.0	45.30		
73	Standard		37011.0	36986.0	453.00		
74	Standard		37233.0	37208.0	453.00		
75	Standard		36837.0	36812.0	453.00		

Buffer Experiment: B1
Experiment Dates 9/30/92 to 10/2/92

Objective: Determine the effect of pH on sorption.

Optical Density: 175 T#3: 0.69

#	Bact.	Buffer	pH	pH Ave. and St. Dev.	CPM/mL	CPM/mL - Background	Initial Conc. (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
31	No	No	8.16	8.18	215.0	181.9	10.00	0.13	19.62
32	No	No	8.12	0.08	196.3	163.2	10.00	0.12	19.65
33	No	No	8.27		228.7	195.6	10.00	0.14	19.60
34	No	Yes	7.93	7.89	879.4	846.3	10.00	0.60	18.69
35	No	Yes	7.86	0.04	836.4	803.3	10.00	0.57	18.75
36	No	Yes	7.88		903.2	870.1	10.00	0.61	18.65
37	Yes	No	7.07	7.09	2332.8	2299.7	10.00	1.62	16.64
38	Yes	No	7.14	0.04	2092.1	2059.0	10.00	1.45	16.98
39	Yes	No	7.06		2315.9	2282.8	10.00	1.61	16.66
40	Yes	Yes	7.82	7.79	889.3	856.2	10.00	0.60	18.67
41	Yes	Yes	7.80	0.04	796.2	763.1	10.00	0.54	18.80
42	Yes	Yes	7.75		869.3	836.2	10.00	0.59	18.70
43	CT Standard		8.06	7.72	28480.8	28447.7	20.00		
44	CT Standard		7.47	0.31	27898.0	27864.9	20.00		
45	CT Standard		7.63		28335.4	28302.3	20.00		

Ave. pH with buffer: 7.84
s of pH with buffer: 0.06
Note: Sample numbers started with 31 for this experiments.

Appendix C

Cd Batch Equilibrium Experiments

APPENDIX C

CADMIUM BATCH EQUILIBRIUM EXPERIMENTS

This section contains data from two experiments performed to determine the effect of microbes on Cd^{2+} sorption to crushed tuff (R2 and R3) and one experiment performed to determine when Cd^{2+} sorption to tuff reaches an equilibrium (Cd kinetic). In addition, there is a graph showing the results of the Cd kinetic experiment. Experiment R2 contains both the AA analyses from the New Mexico Bureau of Mines and Mineral Resources (NMBMMR) Chemistry Laboratory and the ICP analyses from the Soil, Plant, and Water (SWAT) laboratory of New Mexico State University. Samples from the NMIMT are replicates of SWAT samples having the same sample number. Figure 4 in Chapter 2 is from the SWAT data. Results from experiment R3 are displayed in Figure 5 of Chapter 2.

I did not use the lower Cd^{2+} concentrations data because the data appeared to be random. I concluded that the chemical analyses were not sensitive enough at low concentrations.

Batch Experiment: R2
Experiment Dates: 6/26/91 to 7/1/91

Objective: Find the effect of 50 T#7 and 175 T#1 on Cd sorption.

Optical Densities:		
	50 T#7:	0.95
	175 T#1:	0.98

Analysis done at the SWAT Laboratory

#	Sample type	Initial Conc. x 2 (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	50 T#7	0.18	<.01	-
2	50 T#7	0.18	<.01	-
3	50 T#7	0.18	<.01	-
4	50 T#7	0.91	<.01	-
5	50 T#7	0.91	<.01	-
6	50 T#7	0.91	<.01	-
7	50 T#7	2.49	0.03	2.43
8	50 T#7	2.49	0.03	2.43
9	50 T#7	2.49	0.02	2.45
10	50 T#7	10.21	No analysis	-
11	50 T#7	10.21	0.12	9.97
12	50 T#7	10.21	0.12	9.97
13	50 T#7	25.46	0.48	24.50
14	50 T#7	25.46	0.39	24.68
15	50 T#7	25.46	0.44	24.58
16	50 T#7	103.58	3.69	96.20
17	50 T#7	103.58	3.41	96.76
18	50 T#7	103.58	3.43	96.72
19	50 T#7	253.08	14.69	223.70
20	50 T#7	253.08	15.91	221.26
21	50 T#7	253.08	14.7	223.68
22	175 T#1	0.18	0.02	0.14
23	175 T#1	0.18	<.01	-
24	175 T#1	0.18	<.01	-
25	175 T#1	0.91	<.01	-
26	175 T#1	0.91	<.01	-
27	175 T#1	0.91	0.05	0.81
28	175 T#1	2.49	0.05	2.39
29	175 T#1	2.49	0.2	Not used
30	175 T#1	2.49	0.04	2.41
31	175 T#1	10.21	0.15	9.91
32	175 T#1	10.21	0.12	9.97
33	175 T#1	10.21	0.12	9.97
34	175 T#1	25.46	0.45	24.56
35	175 T#1	25.46	0.38	24.70
36	175 T#1	25.46	0.45	24.56
37	175 T#1	103.58	3.12	97.34
38	175 T#1	103.58	3.61	96.36
39	175 T#1	103.58	3.6	96.38

Batch Experiment: R2
Experiment Dates: 6/26/91 to 7/1/91

Objective: Find the effect of 50 T#7 and 175 T#1 on Cd sorption.

Optical Densities:		
	50 T#7:	0.95
	175 T#1:	0.98

Analysis done at the SWAT Laboratory

#	Sample type	Initial Conc. x 2 (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
40	175 T#1	253.08	14.02	225.04
41	175 T#1	253.08	12.43	228.22
42	175 T#1	253.08	13.1	226.88
43	sterile	0.18	0.01	0.16
44	sterile	0.18	<.01	-
45	sterile	0.18	0.02	0.14
46	sterile	0.91	<.01	-
47	sterile	0.91	0.02	0.87
48	sterile	0.91	0.05	0.81
49	sterile	2.49	<0.01	-
50	sterile	2.49	0.08	2.33
51	sterile	2.49	0.11	2.27
52	sterile	10.21	0.29	9.63
53	sterile	10.21	0.11	9.99
54	sterile	10.21	0.02	10.17
55	sterile	25.46	0.05	25.36
56	sterile	25.46	0.05	25.36
57	sterile	25.46	0.04	25.38
58	sterile	103.58	0.6	102.38
59	sterile	103.58	0.57	102.44
60	sterile	103.58	0.58	102.42
61	sterile	253.08	5.38	242.32
62	sterile	253.08	5.71	241.66
63	sterile	253.08	5.48	242.12
64	Standard		0.18	
65	Standard		0.19	
66	Standard		0.18	
67	Standard		0.92	
68	Standard		0.87	
69	Standard		0.94	
70	Standard		2.48	
71	Standard		2.46	
72	Standard		2.52	
73	Standard		10.45	
74	Standard		10.11	
75	Standard		10.08	
76	Standard		25.65	
77	Standard		25.08	
78	Standard		25.64	

Batch Experiment: R2
Experiment Dates: 6/26/91 to 7/1/91

Objective: Find the effect of 50 T#7 and 175 T#1 on Cd sorption.

Optical Densities:		
	50 T#7:	0.95
	175 T#1:	0.98

Analysis done at the SWAT Laboratory

#	Sample type	Initial Conc. x 2 (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
79	Standard		107.12	
80	Standard		101.34	
81	Standard		102.29	
82	Standard		259.77	
83	Standard		250.49	
84	Standard		248.99	

Batch Experiment: R2
Experiment Dates: 6/26/91 to 7/1/91

Objective: Find the effect of 50 T#7 and 175 T#1 on Cd sorption.

Data from the NMBMMR Chemistry Laboratory:

#	Sample type	Initial Conc. x 2 (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
3A	50 T#7	0.1	<.05	-
6A	50 T#7	0.8	<.05	-
8A	50 T#7	2.2	<.05	-
10	50 T#7	9.2	0.1	9
13A	50 T#7	23	0.4	22.2
16A	50 T#7	90	3	84
20A	50 T#7	220	12.2	195.6
23A	175 T#1	0.1	<.05	-
26A	175 T#1	0.8	<.05	-
30A	175 T#1	2.2	<.05	-
32A	175 T#1	9.2	0.09	9.02
34A	175 T#1	23	0.3	22.4
39A	175 T#1	90	2.6	84.8
42A	175 T#1	220	10.3	199.4
45A	sterile	0.1	<.05	-
48A	sterile	0.8	<.05	-
50A	sterile	2.2	<.05	-
54A	sterile	9.2	<.05	-
56A	sterile	23	<.05	-
58A	sterile	90	0.4	89.2
63A	sterile	220	5.2	209.6
64A	Standard		0.1	
67A	Standard		0.8	
70A	Standard		2.2	
73A	Standard		9.2	
76A	Standard		23	
79A	Standard		90	
82A	Standard		220	

Batch Experiment: R3
Experiment Dates: 8/5/91 to 8/7/91

Objective: Find the effect of 50 T#12 and 175 T#3 on Cd sorption to tuff.

Optical Densities: 50 T#12: 1.80
175 T#3: 0.85

Analysis done at the SWAT Laboratory

#	Sample type	Initial Conc. x 2 (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
1	50 T#12	0.16	0.01	0.14
2	50 T#12	0.16	<.01	-
3	50 T#12	0.16	<.01	-
4	50 T#12	1.87	0.02	1.83
5	50 T#12	1.87	0.02	1.83
6	50 T#12	1.87	0.02	1.83
7	50 T#12	9.88	0.16	9.56
8	50 T#12	9.88	0.15	9.58
9	50 T#12	9.88	0.16	9.56
10	50 T#12	25.97	0.59	24.79
11	50 T#12	25.97	0.61	24.75
12	50 T#12	25.97	0.66	24.65
13	50 T#12	248.76	19.88	209.00
14	50 T#12	248.76	19.26	210.24
15	50 T#12	248.76	17.60	213.56
16	175 T#3	0.16	<.01	-
17	175 T#3	0.16	0.01	0.14
18	175 T#3	0.16	0.01	0.14
19	175 T#3	1.87	0.03	1.81
20	175 T#3	1.87	0.03	1.81
21	175 T#3	1.87	0.03	1.81
22	175 T#3	9.88	0.18	9.52
23	175 T#3	9.88	0.22	9.44
24	175 T#3	9.88	0.20	9.48
25	175 T#3	25.97	0.66	24.65
26	175 T#3	25.97	0.63	24.71
27	175 T#3	25.97	0.63	24.71
28	175 T#3	248.76	19.64	209.48
29	175 T#3	248.76	19.83	209.10
30	175 T#3	248.76	19.45	209.86
31	sterile	0.16	0.02	0.12
32	sterile	0.16	<.01	-
33	sterile	0.16	<.01	-
34	sterile	1.87	<.01	-
35	sterile	1.87	<.01	-
36	sterile	1.87	<.01	-
37	sterile	9.88	0.02	9.84
38	sterile	9.88	0.02	9.84
39	sterile	9.88	0.03	9.82

Batch Experiment: R3
Experiment Dates: 8/5/91 to 8/7/91

Objective: Find the effect of 50 T#12 and 175 T#3 on Cd sorption to tuff.

Optical Densities: 50 T#12: 1.80
175 T#3: 0.85

Analysis done at the SWAT Laboratory

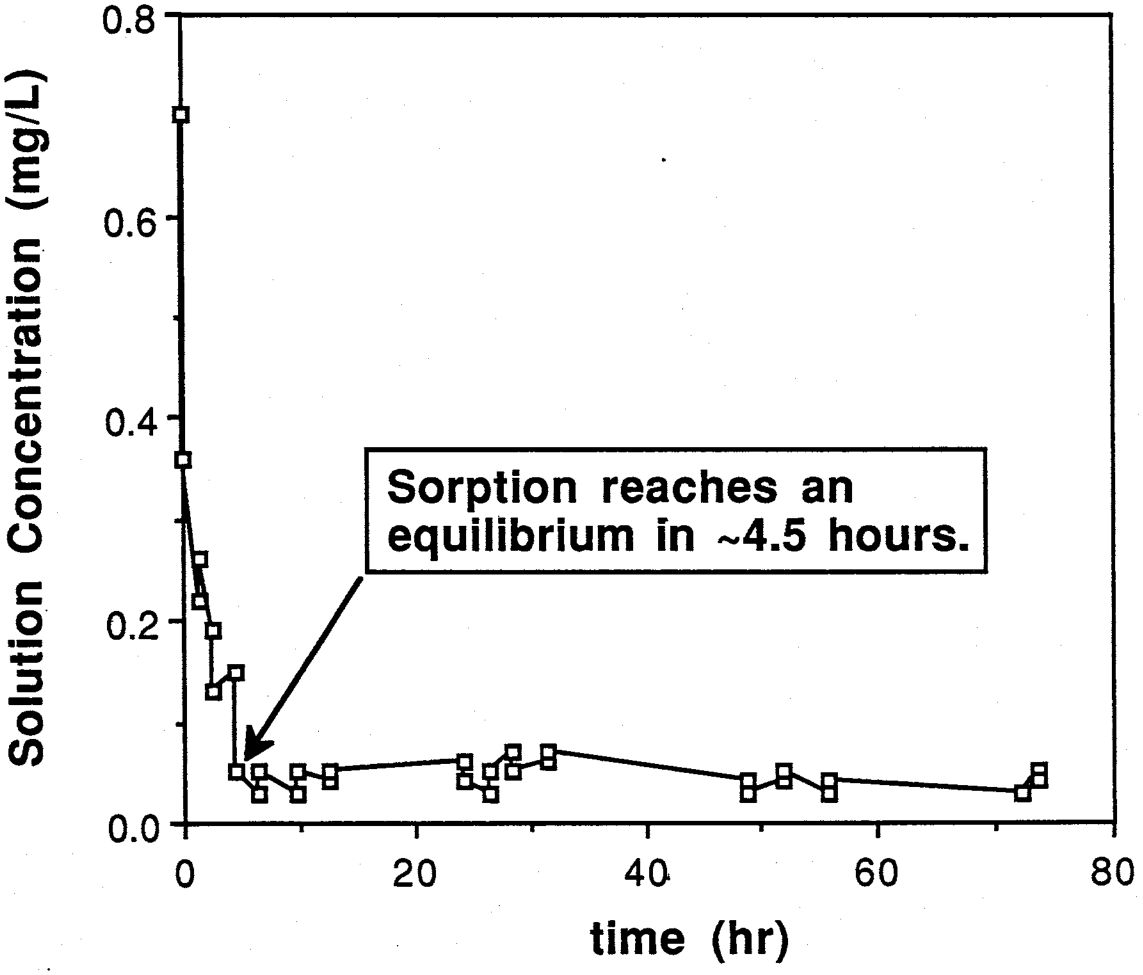
#	Sample type	Initial Conc. x 2 (mg/L)	Sample Conc. (mg/L)	Sorbed Amount (mg/kg)
40	sterile	25.97	0.05	25.87
41	sterile	25.97	0.05	25.87
42	sterile	25.97	0.05	25.87
43	sterile	248.76	6.93	234.90
44	sterile	248.76	6.56	235.64
45	sterile	248.76	7.04	234.68
46	Standard		0.19	
47	Standard		0.15	
48	Standard		0.14	
49	Standard		1.83	
50	Standard		1.90	
51	Standard		1.87	
52	Standard		9.62	
53	Standard		10.02	
54	Standard		9.99	
55	Standard		25.71	
56	Standard		25.70	
57	Standard		26.51	
58	Standard		243.83	
59	Standard		252.19	
60	Standard		250.27	

Experiment: Cd kinetic
Experiment Dates: 7/9/92 to 7/12/91

Objective: Determine how fast Cd in solution reaches an equilibrium with Cd sorbed to crushed tuff.

#	Time (hr)	Sample Conc. C (mg/L)	Amount Sorbed S (mg/kg)
1	0.1	0.7	48.40
2	0.1	0.36	49.08
3	1.3	0.22	49.36
4	1.3	0.26	49.28
5	2.5	0.19	49.42
6	2.5	0.13	49.54
7	4.5	0.15	49.50
8	4.5	0.05	49.70
9	6.5	0.03	49.74
10	6.5	0.05	49.70
11	9.8	0.03	49.74
12	9.8	0.05	49.70
13	12.5	0.04	49.72
14	12.5	0.05	49.70
15	24.3	0.06	49.68
16	24.3	0.04	49.72
17	26.5	0.03	49.74
18	26.5	0.05	49.70
19	28.5	0.07	49.66
20	28.5	0.05	49.70
21	31.5	0.06	49.68
22	31.5	0.07	49.66
23	48.8	0.04	49.72
24	48.8	0.03	49.74
25	51.8	0.04	49.72
26	51.8	0.05	49.70
27	55.7	0.03	49.74
28	55.7	0.04	49.72
29	72.3	0.03	49.74
30	72.3	0.03	49.74
31	73.5	0.05	49.70
32	73.5	0.04	49.72
33	Standard	24.74	
34	Standard	25.06	

Experiment: Cd kinetic



Sorption reaches an equilibrium in ~4.5 hours.

Appendix D

Transport Experiment

APPENDIX D

TRANSPORT EXPERIMENT

This appendix contains the results of a transport experiment designated as 'C2' in my laboratory notebook and results of a saturated conductivity test (D1) carried out on columns packed separately from the columns packed for 'C2'. Information from 'C2' include the effluent flux (D2), dead volume in the apparatus (D3), pore volumes determination (D4), liquid scintillation counts from influent solutions (D5), atomic adsorption analyses of Ni influent solutions (D6), Br^- effluent concentrations (D7), Ni^{2+} effluent concentrations (D8), and the column effluent pH measurements (D9). I have not presented any parameters determined by fitting the BTCs to the CXTFIT two-region model because the optimization of the Br^- BTCs did not converge and there were too many variables (with the addition of R) for the optimization of Ni^{2+} BTCs to converge to reasonable parameter values.

Results from the saturated conductivity tests (D1) are reported in the Transport Experiment methods section of Chapter 2.

The section on effluent flux (D2) also contains both the date and days from the beginning of the experiment at which the columns had a certain flux. Fluxes were calculated by measuring the column effluent volume accumulated over time.

Section D3 contains information on the volume of water within the apparatus (dead volume). The dead volume includes volume in connecting tubing, porous endplates, and the endspace existing in the top and bottom of the columns. An illustration in section D3 shows the location and dimensions of these parts of the apparatus.

Section D4 contains information on the volume of water (one pore volume) contained in each column at the end of the experiment and on the dry bulk density of the packed columns. The volume of water was found by subtracting the dried column apparatus mass and the oven-dried tuff mass from the mass of the wet packed column. I used the average volume from all four columns to calculate the number of pore volumes passing through each individual column. Reasons for this include (1) I believe that the volume of water may have changed during the experiment due to trapped air, since I did not used deaired water and did not saturate the columns with CO_2 prior to starting the experiment and (2) the columns were all packed in the same manner. Upon reviewing my results, I now believe that a better method to calculate pore volumes is to use the measured volume in each column. This would result in different CXTFIT optimized parameters. However, the trends for the fitted parameters would be the same as those using the average pore volumes; therefore the discussion found in chapter 2 would not change.

Section D5 contains information on liquid scintillation count of the influent solution. I used 20 bottles of influent solution for columns 1 and 2 (sterile) and 12 bottles for columns 3 and 4 (inoculated). I measured the LSC of the solution within the bottles before attaching them to the columns (notated 'before' in the appendix) and the solution left over in the bottom of the bottle after removing the solution from the columns (notated 'after' in the appendix).

To make the influent solution, I first made up a stock solution containing approximately 1000 mg L^{-1} Ni^{2+} labeled with ^{63}Ni. This solution was autoclaved and kept in sealed containers. I marked the 6 L line on the influent bottles, so that after autoclaving solutions containing $CaCl_2$, I could add 60 mL of the stock solution, then fill the bottles up to the 6 L line with NANOpure water. To prepare the solutions for the sterile columns, I autoclaved concentrated solutions of NaN_3 and added them to the influent bottles prior to filling with NANOpure water because when Ni^{2+} and NaN3 are heated together, a precipitate forms. I used disconnects between tubing to prevent microbial contamination when I changed the solutions.

Section D6 shows the results of atomic adsorption analyses of Ni^{2+} influent solution concentrations. This data is not extensive, so my conclusion that the average influent solution concentration is 9.3 mg/L is probably not correct; however, the actual concentration of the influent solution does not affect the conclusions, since I used relative concentrations (C/Co) for parameter fitting of the Ni^{2+} BTCs.

Bromide effluent concentrations are given in section D7 of this appendix along with the high performance liquid chromatography (HPLC) peak heights used to calculate these concentrations. The Br^- concentration of the effluent prior to adding Br^- to the columns and for the influent solutions to each column is also given. The actual volume of each sample was not measured, but I did measure the flux before adding the slug, after taking samples for the upward limb, and after the slug finished. Therefore, to calculate PV's, I used the flux measured before the columns were attached for the upward limb of the BTCs and the flux measured after Br^- slug finished for the downward limb of the BTCs. Note that to optimize the velocity, I needed to use time instead of PVs. Since the flux changed during the time I added Br^-, I recalculated time at a constant flux.

The flux I used for this was the same as the measured flux during the middle part of the breakthrough curve. After determining the PV's for each column, I subtracted 0.3 PV to account for dead volume.

Nickel effluent concentrations are given in section D8. The dead volume was subtracted from the number of pore volumes even though the amount was negligible. C/Co was calculated by dividing the sample CPM by the average standard CPM after background counts were subtracted from each value.

The pH values of the column influent and effluent solutions are given in section D9.

SECTION D1
SATURATED CONDUCTIVITY TESTS

The following tests were performed on unautoclaved tuff.
The item being tested was saturated with CO2 before the test begun and the water was deaired.
The water in the falling head permeameter test was run from the top to the bottom of the columns.

Date	Hydraulic Conductivity (m/sec)	Comment
12/22/91	4.80E-07	Conductivity of the frit
12/22/91	3.10E-06	Conductivity of column packed without frits
12/23/91	2.80E-06	Conductivity of 1st column packed with frit
12/27/91	1.80E-06	Conductivity of 2nd column packed with frit

The following tests were performed on a column packed with autoclaved tuff.
The column was saturated with CO2 before the test begun and the water was deaired.

Date	Hydraulic Conductivity (m/sec)	Comment
3/21/92	7.70E-06	With frit, Made sure air was bled from the top using a syringe.
3/22/92	4.10E-06	Same column, Lower K with time:
3/24/92	2.50E-06	Due to compaction? Note large pores or bubbles appear near the top of the column.

SECTION D2
EFFLUENT FLUX

Dates: 7/7/92 to 9/11/93

I used the following to calculate the number of PV's passing through the columns:

Day 0 to day 3: Used the average flux of 0.42 mL/min for all columns.
Day 3 to day 15: Used the average flux of 0.42 mL/min for all columns.
except day 5, when, from 5.17 to 5.96 days, only 105.5 mL passed through column 3 and 73.8 mL passed through column 4.
Day 15 to day 34: Used the average flux of 0.42 mL/min for all columns.
Day 34 to end: Due to variability in pumping, used the fluxes listed.

		Column fluxes in mL/min			
Date	Days from start	Column 1	Column 2	Column 3	Column 4
7/6/92	-1	0.42	0.42	0.43	0.43
7/6/92	-1	0.41	0.42	0.42	0.43
7/10/92	3	0.41	0.40	0.41	0.40
7/11 to 7/12	3.25 to 4.75	0.21	0.21	0.21	0.20
7/12 to 7/14	5.2 to 7.5	0.21	0.21	0.21	0.21
7/16 to 7/21	9.1 to 14.5	0.20	0.21	0.21	0.20
7/22 to 7/23	15 to 16	0.40	0.41	0.41	0.40
7/23 to 7/24	16 to 17	0.42	0.42	0.42	0.42
7/26 to 7/27	19 to 20	0.41	0.42	0.43	0.41
7/27 to 7.28	20 to 21	0.42	0.42	0.41	0.40
7/29 to 7/30	21 to 22	0.41	0.42	0.41	0.40
7/31 to 8/1	23 to 24	0.43	0.41	0.41	0.41
8/1 to 8/2	24 to 25	0.42	0.41	0.41	0.42
8/4 to 8/5	28 to 29	0.40	0.40	0.41	0.40
8/9 to 8/10	33 to 34	0.41	0.40	0.41	0.40
8/12 to8/13	36 to 37	0.40	0.39	0.40	0.41
8/17 to 8/18	41 to 42	0.40	0.37	0.40	0.40
8/20 to 8/21	44 to 45	0.39	0.39		
8/22 to 8/23	46 to 47	0.35	0.34		
8/23 to 8/24	47 to 48	0.34	0.38		
8/24 to 8/25	48 to 49	0.38	0.39		
8/25 to 8/26	49 to 50	0.38	0.40		
8/26 to 8/27	50 to -51	0.38	0.38		
8/28 to 8/29	52 to 53	0.38	0.39		
8/29 to 8/30	53 to 54	0.38	0.38		
8/30 to 8/31	54 to 55	0.38	0.38		
8/31 to 9/1	55 to 56	0.36	0.36		
9/1 to 9/2	56 to 57	0.38	0.37		
9/2 to 9/3	57 to 58	0.39	0.39		
9/3 to 9/4	58 to 59	0.33	0.37		
9/4 to 9/5	59 to 60	0.34	0.36		
9/5 to 9/6	60 to 61	0.33	0.36		
9/6 to 9/7	61 to 62	0.32	0.34		
9/7 to 9/8	62 to 63	0.34	0.37		
9/8 to 9/9	63 to 64	0.35	0.38		
9/9 to 9/10	64 to 65	0.34	0.38		
9/10 to 9/11	65 to 66	0.35	0.38		

SECTION D3
COLUMN APPARATUS VOLUME

Date: 10/2/92

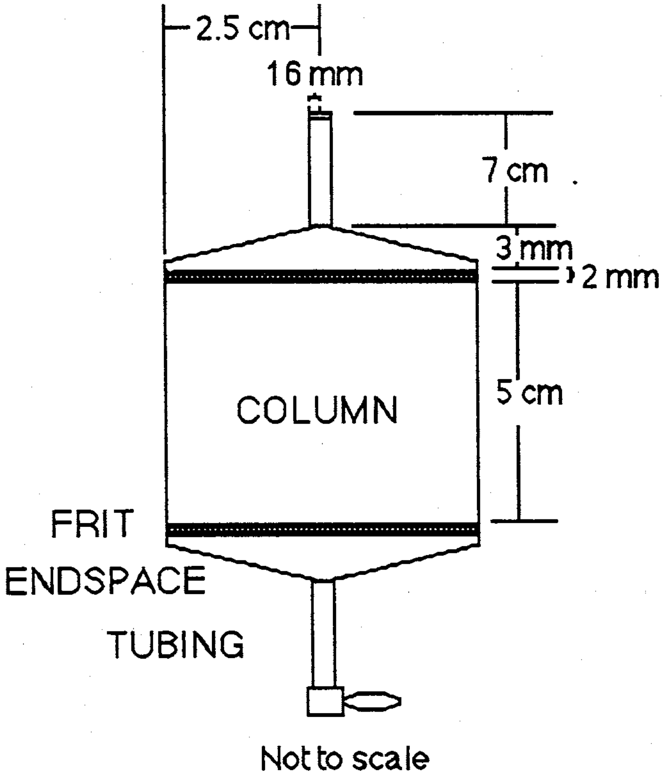
Method: To determine the maximum dead volume in the apparatus, I put the column together without any crushed tuff, then pumped CO_2 through the column.
I deaired a water by attaching the solution to a vacuum, then slowly pumped the water through the column. The wet mass of the column was subtracted from the dry mass to determine the volume of water in the column and in the apparatus, then I subtracted the volume of water in the column from the total volume:

Mass of dry column:	429.9 g
Mass of column and water:	539.0 g
Volume of water:	110.0 mL
Mass of water in column ($\pi \times 2.5^2 \times 5$) mL:	98.2 mL
Mass of water in apparatus:	11.8 mL

Based on the following calculations, a dead space of 11.8 mL is reasonable (note that various parts of the apparatus including the tubing, frit, and endspace are shown in the figure on the next page:

Volume of water in tubing = $2 \times \pi r^2$ = 1.1 mL
Volume of water in endplates = $2 \times (\pi r^2 h + \pi r^2 h/3)$ = 6.5 mL
Volume of water in frits = $2 \times n \times \pi r^2$ = 3.2 mL
(n = frit porosity)
Total = 10.8 mL

SECTION D3
COLUMN APPARATUS VOLUME



Column apparatus showing the deadspace.

SECTION D4
COLUMN PORE VOLUMES, BULK DENSITY

Dates: 7/7/92 to 9/11/92

	Column 1	Column 2	Column 3	Column 4	average	standard deviation
1 Mass of tuff (g):	137.7	136.3	136.9	134.95	136.5	1.2
2 Mass of water in the column + apparatus (g):	51.5	54.4	52.9	48.2	51.8	2.6
3 Pore Volumes (mL) (subtract 11.8 from row 2)	39.7	42.6	41.1	36.4	40.0	2.6

Ave. porosity = average PV/column volume: 0.41
Ave. bulk density = ave. tuff mass/column mass 1.39 g/mL

SECTION D5
NI INFLUENT SOLUTION LIQUID SCINTILLATION COUNTS

Dates: 7/7/92 to 7/25/92

Summary statistics

Universol Cocktale:

Average influent solution CPM/mL:	13053.2
Standard deviation of influent solution CPM/mL:	704
Average background CPM/mL:	37.7
Standard deviation of background CPM/mL:	4.2

Scintiverse Cocktale:

Average influent solution CPM/mL:	13133.9
Standard deviation of influent solution CPM/mL:	610

Using Universol Cocktale:

	first	second	third	fourth	fifth	sixth
Date of influent soln. connection:	7/7/92	7/12/92	7/15/92	7/23/92	7/25/92	7/27-Co1&2 7/29-Co3&4
Col 1 (before)	13682.2	10840.0	13427.5	13337.8		12504.6
Col 1 (after)	Too low	13471.8	13054.3			
Col 2 (before)	12036.1	12939.0	13242.8	12730.5		14918.9
Col 2 (after)	13314.8	13726.9	13321.7			
Col 3 (before)	13187.7	13089.3	13688.4	13174.9		13568.8
Col 3 (after)	11364.5	12947.0	11856.1		13549.00	
Col 4 (before)	12510.7	13093.5	13300.7	13260.2		14313.8
Col 4 (after)	12610.7	13138.7	13210.5		13586.40	

SECTION D5
NI INFLUENT SOLUTION LIQUID SCINTILLATION COUNTS

Dates: 7/7/92 to 7/25/92

Using Scintiverse Cocktale:

	4th	5th*	6th	7th	8th
Date of infl soln connection:	7/23/92	7/25/92	7/27-Co1&2 7/29-Co 3&	7/31/92	8/2/92
Col 1 (before)		11627.9		14490.9 too high	
Col 1 (after)	13013.0	12716.5	13037.7	13704.0	14524.30
Col 2 (before)		11292.9		13530.0 too high	
Col 2 (after)	13139.8	12471.9	13439.7	13240.3	
Col 3 (before)		12520.6		13683.9 too high	
Col 3 (after)	13055.7	13400.5	14086.6	13542.6	14745.80
Col 4 (before)		12765.0		13498.5 too high	
Col 4 (after)	12875.0	13231.6	14176.8	12895.5	

	9th	10th	11th	12th	13th	14th
Date of infl soln connection:	8/6/92	8/8/92	8/12/92	8/14/92	8/19/92?	8/24/1992?
Col 1&2 (before)	13381.2	13582.8	13224.6	13157.0	14788.1	13678.4
Col 1&2 (after)	10039.4	13366.7	13331.2	13566.3	13044.1	12934.2
Col 3&4 (before)	13598.2	13183.8	13415.3	13756.4		
Col 3&4 (after)	12170.9	13590.7	13308.0	13614.4		

	15th	16th	17th	18th	19th	20th
Date of infl soln connection:	8/28/92	9/2/92	9/7/92	9/12/92	9/17/92	9/22/92
Col 1&2 (before)	13120.1	13938.3	13045.1	11924.1	13069.1	13485.9
Col 1&2 (after)	12866.4	13391.1	12894.4	12675.6	14093.3	

* The 5th influent soln contained colloids or suspended particals:
When mixed, the samples showed a higher count, possibly due to the particals in suspension which could either be molecules containing Nickel or molecules with sorbed Ni.
This solution was warm when the Ni was mixed in.

SECTION D6
ATOMIC ADSORPTION ANALYSES ON COLUMN INFLUENT SOLUTIONS

Stock Solution	Concentration	ave.	ave. of last 3 (No LSC done)
# 1	9.76	9.42	9.31
# 2	9.20		
# 3	9.36		
# 4	9.37		

The following table compares results of LSC analysis verses AA
if C is calculated from LSC using C = 9.31 when CPM/mL is 13100 CF

Sample	CPM	C (mg/L) (from CPM)	C (mg/L) AA	difference
1st, 7/7 Col1	13682.2	9.72	11.05	-1.3262
1st, 7/12 Col1	8321.5	5.91	5.41	0.5040
3rd 7/23 Col 1	13054.3	9.28	8.04	1.2375
4th 7/23 Col4	13260.2	9.42	8.66	0.7639
6th 7/31 Col3	14086.6	10.01	9.12	0.8912
8th 8/2 Col 1	20986.0	14.91	14.57	0.3445
8th 8/6 Col 1&2	14524.3	10.32	9.45	0.8722
6th,7/27,col1	12504.6	8.89	8.72	0.1669
6th,7/29,Col3	13568.8	9.64	10.68	-1.0368

SECTION D7
BROMIDE EFFLUENT CONCENTRATIONS

Experiment Dates: 7/31/92 to 8/2/92

BR DATA, Analyzed using the HPLC
HPLC Br standard concentration = 20 mg/L

Solution Type	HPLC sample reading	HPLC standard reading	Sample Conc. (mg/L)
Column 1 effluent before Br	0	129072	0.00
Column 3 effluent before Br	325	144692	0.04
Column 1&2 influent solutior	77976	184104	8.47
Column 1&2 influent solutior	78472	184104	8.52
Column 3&4 influent solutior	54091	144692	7.48
Column 3&4 influent solutior	53715	144692	7.42

Column 1

time (hr)	HPLC sample reading	HPLC standard reading	Sample Conc. (mg/L)	Relative Conc. C/Co	Pore Volumes	Pore Vol. - Dead Volume	Calc. time (d)
0.4	0	184104	0.00	0.00	0.25	-0.04	0.00
0.6	0	184104	0.00	0.00	0.38	0.08	0.01
0.8	10392	184104	1.13	0.13	0.50	0.21	0.01
1.0	13653	184104	1.48	0.17	0.63	0.34	0.02
1.2	17109	184104	1.86	0.22	0.76	0.46	0.03
1.4	21555	184104	2.34	0.28	0.88	0.59	0.04
1.8	31709	183384	3.46	0.41	1.14	0.84	0.05
2.2	43933	183384	4.79	0.56	1.39	1.09	0.07
2.6	55421	183384	6.04	0.71	1.64	1.34	0.09
3.0	66661	183384	7.27	0.86	1.89	1.60	0.10
48.0	79421	183384	8.66	1.02	30.95	30.66	2.00
48.4	79333	183384	8.65	1.02	31.21	30.91	2.01
48.8	74768	183384	8.15	0.96	31.46	31.17	2.03
49.2	69224	183384	7.55	0.89	31.72	31.42	2.05
49.6	60632	183384	6.61	0.78	31.97	31.68	2.06
50.0	49027	183384	5.35	0.63	32.23	31.93	2.08
50.4	35299	184323	3.83	0.45	32.49	32.19	2.10
50.8	20456	184323	2.22	0.26	32.74	32.45	2.11
51.2	4304	184323	0.47	0.05	33.00	32.70	2.13
51.6	1595	144692	0.22	0.03	33.25	32.96	2.15
52.0	0	144692	0.00	0.00	33.51	33.21	2.16
52.4	789	144692	0.11	0.01	33.76	33.47	2.18
52.8	931	144692	0.13	0.02	34.02	33.72	2.20

SECTION D7
BROMIDE EFFLUENT CONCENTRATIONS

Experiment Dates: 7/31/92 to 8/2/92

BR DATA, Analyzed using the HPLC
HPLC Br standard concentration = 20 mg/L

Column 2

time (hr)	HPLC sample reading	HPLC standard reading	Sample Conc. (mg/L)	Relative Conc. C/Co	Pore Volumes	Pore Vol. - Dead Volume	Calc. time (d)
0.2	0	125003	0.00	0.00	0.12	-0.17	-0.01
0.6	2616	125003	0.42	0.05	0.37	0.08	0.01
1.0	10525	125003	1.68	0.20	0.62	0.33	0.02
1.4	17544	125003	2.81	0.33	0.87	0.58	0.04
1.8	23051	125003	3.69	0.43	1.12	0.83	0.06
2.2	28709	125003	4.59	0.54	1.37	1.08	0.07
2.6	33691	125003	5.39	0.63	1.62	1.33	0.09
3.0	38360	125003	6.14	0.72	1.87	1.57	0.11
48.2	50472	125003	8.08	0.95	29.70	29.41	1.98
48.6	50056	125003	8.01	0.94	29.95	29.65	1.99
49.0	42152	123808	6.81	0.80	30.20	29.90	2.01
49.4	35315	123808	5.70	0.67	30.44	30.15	2.03
49.8	29221	123808	4.72	0.56	30.69	30.39	2.04
50.2	23528	123808	3.80	0.45	30.93	30.64	2.06
50.6	18401	123808	2.97	0.35	31.18	30.89	2.08
51.0	13571	123808	2.19	0.26	31.43	31.13	2.09
51.4	9672	123808	1.56	0.18	31.67	31.38	2.11
51.8	6896	123808	1.11	0.13	31.92	31.62	2.13
52.2	4728	123808	0.76	0.09	32.17	31.87	2.14
52.6	2592	123808	0.42	0.05	32.41	32.12	2.16
53.0	2241	123787	0.36	0.04	32.66	32.36	2.17

SECTION D7
BROMIDE EFFLUENT CONCENTRATIONS

Experiment Dates: 7/31/92 to 8/2/92

BR DATA, Analyzed using the HPLC
HPLC Br standard concentration = 20 mg/L

Column 3

time (hr)	HPLC sample reading	HPLC standard reading	Sample Conc. (mg/L)	Relative Conc. C/Co	Pore Volumes	Pore Vol. - Dead Volume	Calc. time (d)
0.0	0	184323	0.00	0.00	0.00	-0.30	-0.02
0.2	0	185759	0.00	0.00	0.12	-0.17	-0.01
0.4	0	185759	0.00	0.00	0.25	-0.05	0.00
0.6	0	185759	0.00	0.00	0.37	0.07	0.01
0.8	0	185759	0.00	0.00	0.49	0.20	0.01
1.0	0	144692	0.00	0.00	0.62	0.32	0.02
1.4	1333	144692	0.18	0.02	0.86	0.57	0.04
1.8	24363	185759	2.62	0.35	1.11	0.81	0.05
2.0	37440	185759	4.03	0.54	1.23	0.94	0.06
2.2	34245	144692	4.73	0.64	1.35	1.06	0.07
2.4	36121	125184	5.77	0.77	1.48	1.18	0.08
2.6	39643	125184	6.33	0.85	1.60	1.31	0.09
2.8	41909	125184	6.70	0.90	1.72	1.43	0.10
3.0	43752	125184	6.99	0.94	1.85	1.55	0.10
49.2	47605	125184	7.61	1.02	30.30	30.00	2.03
49.4	47393	125184	7.57	1.02	30.42	30.12	2.03
49.8	42805	125184	6.84	0.92	30.67	30.37	2.05
50.0	34593	125184	5.53	0.74	30.79	30.49	2.06
50.2	25485	125184	4.07	0.55	30.91	30.62	2.07
50.6	12688	125184	2.03	0.27	31.16	30.86	2.08
51.0	6373	125184	1.02	0.14	31.40	31.11	2.10
51.4	2645	125184	0.42	0.06	31.65	31.36	2.12
51.8	1395	125184	0.22	0.03	31.90	31.60	2.13
52.0	1141	125184	0.18	0.02	32.02	31.72	2.14
52.2	1128	125184	0.18	0.02	32.14	31.85	2.15
52.4	16	125184	0.00	0.00	32.27	31.97	2.16
52.6	0	125184	0.00	0.00	32.39	32.09	2.17
52.8	0	125184	0.00	0.00	32.51	32.22	2.17

SECTION D7
BROMIDE EFFLUENT CONCENTRATIONS

Experiment Dates: 7/31/92 to 8/2/92

BR DATA, Analyzed using the HPLC
HPLC Br standard concentration = 20 mg/L

Column 4

time (hr)	HPLC sample reading	HPLC standard reading	Sample Conc. (mg/L)	Relative Conc. C/Co	Pore Volumes	Pore Vol. - Dead Volume	Calc. time (d)
1.0	0	123787	0.00	0.00	0.61	0.31	0.02
1.4	867	123787	0.14	0.02	0.85	0.56	0.04
1.8	17245	123787	2.79	0.37	1.09	0.80	0.05
2.0	29185	123787	4.72	0.63	1.22	0.92	0.06
2.2	35707	123787	5.77	0.77	1.34	1.04	0.07
2.4	39768	123787	6.43	0.86	1.46	1.16	0.08
2.6	41645	123787	6.73	0.90	1.58	1.29	0.09
2.8	43443	123787	7.02	0.94	1.70	1.41	0.09
3.0	44333	123787	7.16	0.96	1.82	1.53	0.10
48.8	48736	128037	7.61	1.02	30.03	29.74	1.99
49.0	49168	128037	7.68	1.03	30.16	29.86	2.00
49.2	49647	128037	7.76	1.04	30.28	29.99	2.01
49.8	46088	128037	7.20	0.97	30.66	30.36	2.03
50.0	37584	128037	5.87	0.79	30.78	30.49	2.04
50.2	25267	128037	3.95	0.53	30.91	30.61	2.05
50.4	49403	128037	7.72	1.04	31.03	30.73	
50.6	8315	128037	1.30	0.17	31.15	30.86	2.06
51.0	3451	128037	0.54	0.07	31.40	31.11	2.08
51.4	1837	128037	0.29	0.04	31.65	31.36	2.10
51.8	656	128445	0.10	0.01	31.90	31.61	2.11
52.0	536	128445	0.08	0.01	32.03	31.73	2.12
52.2	0	128445	0.00	0.00	32.15	31.86	2.13

SECTION D8
NICKEL EFFLUENT CONCENTRATIONS

Dates: 7/7/92 to 9/25/92

Column 1

Time (d)	Sample LSC (CPM/mL)	Sample - Background (CPM/mL)	Pore Volumes	Pore Vol. - Dead Vol.	Conc.	Relative Conc.
0.00	35.8	-1.9	0.06	-0.23	0.00	0.00
1.00	36.5	-1.2	15.14	14.84	0.00	0.00
2.00	43.4	5.7	30.28	29.98	0.00	0.00
3.00	33.4	-4.3	45.42	45.12	0.00	0.00
4.00	47.6	9.9	54.88	54.58	0.01	0.00
4.50	182.4	144.7	58.66	58.37	0.10	0.01
5.00	1206.7	1169.0	62.45	62.15	0.83	0.09
5.50	775.3	737.6	66.23	65.94	0.53	0.06
6.00	1212.0	1174.3	70.02	69.72	0.84	0.09
7.00	1523.9	1486.2	77.59	77.29	1.06	0.11
7.93	1913.5	1875.8	84.65	84.36	1.34	0.14
8.93	3556.0	3518.3	92.22	91.93	2.51	0.27
9.93	2924.2	2886.5	99.79	99.50	2.06	0.22
10.93	3790.6	3752.9	107.36	107.06	2.68	0.29
11.93	3288.4	3250.7	114.93	114.63	2.32	0.25
12.93	3130.8	3093.1	122.50	122.20	2.21	0.24
13.93	3576.2	3538.5	130.07	129.77	2.52	0.27
14.93	3342.6	3304.9	137.64	137.34	2.36	0.25
15.96	5385.0	5347.3	151.61	151.31	3.81	0.41
16.20	5201.6	5163.9	155.16	154.86	3.68	0.40
17.00	4963.0	4925.3	166.98	166.68	3.51	0.38
17.26	4821.4	4783.7	170.82	170.53	3.41	0.37
18.10	4846.8	4809.1	183.24	182.94	3.43	0.37
19.30	4097.2	4059.5	200.97	200.67	2.90	0.31
19.96	5415.6	5377.9	210.72	210.43	3.84	0.41
20.96	9070.2	9032.5	225.50	225.21	6.44	0.69
21.96	7328.0	7290.3	240.28	239.98	5.20	0.56
22.96	7896.3	7858.6	255.06	254.76	5.57	0.60
23.98	8044.0	8006.3	270.13	269.84	5.68	0.61
24.96	8267.8	8230.1	284.62	284.32	5.83	0.63
25.96	8646.6	8608.9	299.39	299.10	6.10	0.66
27.96	9137.3	9099.6	328.95	328.66	6.45	0.69
28.94	9002.8	8965.1	343.43	343.14	6.35	0.68
29.96	10210.0	10172.3	358.51	358.21	7.21	0.77
30.96	10266.2	10228.5	373.29	372.99	7.25	0.78
31.92	10196.8	10159.1	387.47	387.18	7.20	0.77
33.09	8931.5	8893.8	404.76	404.47	6.30	0.68
33.94	8978.4	8940.7	417.33	417.03	6.34	0.68
34.93	9903.6	9865.9	431.96	431.66	6.99	0.75
35.96	9756.7	9719.0	446.81	446.51	6.89	0.74
36.96	10572.7	10535.0	461.23	460.93	7.47	0.80
37.96	10311.2	10273.5	475.64	475.35	7.28	0.78
39.06	10205.6	10167.9	491.50	491.21	7.21	0.77

SECTION D8
NICKEL EFFLUENT CONCENTRATIONS

Dates: 7/7/92 to 9/25/92

Column 1

Time (d)	Sample LSC (CPM/mL)	Sample - Background (CPM/mL)	Pore Volumes	Pore Vol. - Dead Vol.	Conc.	Relative Conc.
39.97	11079.7	11042.0	504.62	504.33	7.83	0.84
40.95	11492.3	11454.6	518.75	518.46	8.12	0.87
41.96	11379.2	11341.5	533.32	533.02	8.04	0.86
42.96	11634.0	11596.3	547.73	547.44	8.22	0.88
44.02	12143.2	12105.5	562.63	562.34	8.58	0.92
44.94	12240.0	12202.3	575.57	575.27	8.65	0.93
45.96	12590.2	12552.5	589.24	588.95	8.90	0.96
47.00	12176.2	12138.5	602.36	602.07	8.60	0.92
47.93	12024.6	11986.9	613.76	613.47	8.50	0.91
48.96	12226.2	12188.5	627.87	627.57	8.64	0.93
49.95	11622.5	11584.8	641.43	641.13	8.21	0.88
50.96	11358.3	11320.6	655.26	654.97	8.02	0.86
51.96	11597.3	11559.6	668.96	668.66	8.19	0.88
52.92	11124.2	11086.5	682.11	681.81	7.86	0.84
54.08	10772.1	10734.4	698.00	697.70	7.61	0.82
54.96	11374.9	11337.2	710.05	709.76	8.04	0.86
55.97	11311.3	11273.6	723.16	722.86	7.99	0.86
56.93	11518.5	11480.8	736.31	736.01	8.14	0.87
57.97	11678.3	11640.6	750.93	750.63	8.25	0.89
58.94	11962.7	11925.0	762.46	762.17	8.45	0.91
59.95	11337.6	11299.9	774.84	774.55	8.01	0.86
60.95	11327.0	11289.3	786.74	786.44	8.00	0.86
61.97	11120.4	11082.7	798.50	798.21	7.86	0.84
62.96	9750.0	9712.3	810.64	810.34	6.88	0.74
63.95	11056.8	11019.1	823.12	822.83	7.81	0.84
64.96	11068.0	11030.3	835.50	835.21	7.82	0.84
65.95	11214.9	11177.2	847.99	847.70	7.92	0.85
66.95	11333.1	11295.4			8.01	0.86
68.00	11430.7	11393.0			8.08	0.87
69.00	11279.0	11241.3			7.97	0.86
70.00	11232.8	11195.1			7.94	0.85
72.00	11315.2	11277.5			7.99	0.86
72.90	11911.8	11874.1			8.42	0.90
74.00	12230.3	12192.6			8.64	0.93
75.00	12528.4	12490.7			8.85	0.95
75.90	12793.3	12755.6			9.04	0.97
76.90	12341.6	12303.9			8.72	0.94

SECTION D8
NICKEL EFFLUENT CONCENTRATIONS

Dates: 7/7/92 to 9/25/92

Column 2

Time (d)	Sample LSC (CPM/mL)	Sample - Background (CPM/mL)	Pore Volumes	Pore Vol. - Dead Vol.	Conc.	Relative Conc.
0.00	32.4	-5.3	0.06	-0.23	0.00	0.00
1.00	35.1	-2.6	15.14	14.84	0.00	0.00
2.00	43.4	5.7	30.28	29.98	0.00	0.00
3.00	57.8	20.1	45.42	45.12	0.01	0.00
4.00	212.3	174.6	54.88	54.58	0.12	0.01
4.50	348.7	311.0	58.66	58.37	0.22	0.02
5.00	753.4	715.7	62.45	62.15	0.51	0.05
5.50	484.3	446.6	66.23	65.94	0.32	0.03
6.00	735.2	697.5	70.02	69.72	0.50	0.05
7.00	1191.8	1154.1	77.59	77.29	0.82	0.09
7.93	1414.4	1376.7	84.65	84.36	0.98	0.11
8.93	3386.8	3349.1	92.22	91.93	2.39	0.26
9.93	2245.7	2208.0	99.79	99.50	1.57	0.17
10.93	3103.4	3065.7	107.36	107.06	2.19	0.23
11.93	2577.5	2539.8	114.93	114.63	1.81	0.19
12.93	2512.5	2474.8	122.50	122.20	1.77	0.19
13.93	3131.1	3093.4	130.07	129.77	2.21	0.24
14.93	3039.2	3001.5	137.64	137.34	2.14	0.23
15.96	5482.0	5444.3	145.41	145.11	3.88	0.42
16.20	5731.4	5693.7	155.16	154.86	4.06	0.44
17.00	5732.2	5694.5	166.98	166.68	4.06	0.44
17.26	5599.8	5562.1	170.82	170.53	3.97	0.43
18.10	5269.1	5231.4	183.24	182.94	3.73	0.40
19.30	4777.5	4739.8	200.97	200.67	3.38	0.36
19.96	5577.1	5539.4	210.72	210.43	3.95	0.42
20.96	8060.3	8022.6	225.50	225.21	5.72	0.61
21.96	6868.2	6830.5	240.28	239.98	4.87	0.52
22.96	7529.5	7491.8	255.06	254.76	5.31	0.57
23.98	7728.8	7691.1	270.13	269.84	5.45	0.59
24.96	8051.6	8013.9	284.62	284.32	5.68	0.61
25.96	8312.7	8275.0	299.39	299.10	5.87	0.63
27.96	8870.7	8833.0	328.95	328.66	6.26	0.67
28.94	8869.4	8831.7	343.43	343.14	6.26	0.67
29.96	9888.4	9850.7	358.51	358.21	6.98	0.75
30.96	10140.6	10102.9	373.29	372.99	7.16	0.77
31.92	10317.3	10279.6	387.47	387.18	7.29	0.78
33.09	9071.7	9034.0	404.76	404.47	6.40	0.69
33.94	9037.6	8999.9	417.33	417.03	6.38	0.69
34.93	9954.1	9916.4	431.96	431.66	7.03	0.76
35.96	9813.5	9775.8	446.44	446.14	6.93	0.74
36.96	10793.9	10756.2	460.49	460.20	7.62	0.82
37.96	10714.0	10676.3	474.55	474.26	7.57	0.81

SECTION D8
NICKEL EFFLUENT CONCENTRATIONS

Dates: 7/7/92 to 9/25/92

Column 2

Time (d)	Sample LSC (CPM/mL)	Sample - Background (CPM/mL)	Pore Volumes	Pore Vol. - Dead Vol.	Conc.	Relative Conc.
39.06	10351.8	10314.1	490.01	489.72	7.31	0.79
39.97	11405.0	11367.3	502.15	501.86	8.06	0.87
40.95	11550.8	11513.1	515.22	514.93	8.16	0.88
41.96	11559.5	11521.8	528.69	528.40	8.17	0.88
42.96	11605.3	11567.6	542.03	541.73	8.20	0.88
44.02	12311.9	12274.2	556.93	556.63	8.70	0.93
44.94	12275.3	12237.6	569.86	569.57	8.67	0.93
45.96	12776.8	12739.1	583.37	583.08	9.03	0.97
47.00	12243.0	12205.3	596.12	595.82	8.65	0.93
47.93	12358.1	12320.4	608.86	608.56	8.73	0.94
48.96	12199.9	12162.2	623.33	623.04	8.62	0.93
49.95	11581.1	11543.4	637.61	637.31	8.18	0.88
50.96	11485.5	11447.8	651.44	651.15	8.11	0.87
51.96	11660.5	11622.8	665.31	665.01	8.24	0.88
52.92	11261.6	11223.9	678.80	678.51	7.96	0.85
54.08	10918.4	10880.7	694.69	694.39	7.71	0.83
54.96	11433.4	11395.7	706.74	706.45	8.08	0.87
55.97	11585.6	11547.9	719.85	719.55	8.19	0.88
56.93	11523.5	11485.8	732.65	732.36	8.14	0.87
57.97	11784.9	11747.2	747.27	746.98	8.33	0.89
58.94	12027.2	11989.5	760.21	759.91	8.50	0.91
59.95	11631.9	11594.2	773.31	773.02	8.22	0.88
60.95	11565.2	11527.5	786.29	786.00	8.17	0.88
61.97	11430.0	11392.3	798.79	798.50	8.08	0.87
62.96	10154.2	10116.5	811.99	811.70	7.17	0.77
63.95	11329.8	11292.1	825.55	825.26	8.00	0.86
64.96	11390.3	11352.6	839.39	839.09	8.05	0.86
65.95	11461.6	11423.9	852.95	852.65	8.10	0.87
66.95	11442.6	11404.9			8.08	0.87
68.00	11452.2	11414.5			8.09	0.87
69.00	11511.8	11474.1			8.13	0.87
70.00	11437.5	11399.8			8.08	0.87
72.00	11500.3	11462.6			8.13	0.87
72.90	12414.3	12376.6			8.77	0.94
74.00	12135.3	12097.6			8.58	0.92
75.00	12609.3	12571.6			8.91	0.96
75.90	12704.0	12666.3			8.98	0.96
76.90	11241.9	11204.2			7.94	0.85

SECTION D8
NICKEL EFFLUENT CONCENTRATIONS

Dates: 7/7/92 to 9/25/92

Column 3

Time (d)	Sample LSC (CPM/mL)	Sample - Background (CPM/mL)	Pore Volumes	Pore Vol. - Dead Vol.	Conc.	Relative Conc.
0.00	49.0	11.3	0.06	-0.23	0.01	0.00
1.00	42.8	5.1	15.14	14.84	0.00	0.00
2.00	34.9	-2.8	30.28	29.98	0.00	0.00
3.00	39.9	2.2	45.42	45.12	0.00	0.00
4.00	43.2	5.5	54.88	54.58	0.00	0.00
5.00	35.4	-2.3	62.45	62.15	0.00	0.00
6.00	42.1	4.4	66.63	66.33	0.00	0.00
7.00	35.4	-2.3	74.21	73.91	0.00	0.00
7.93	37.6	-0.1	81.27	80.98	0.00	0.00
8.93	40.7	3.0	88.84	88.55	0.00	0.00
9.93	43.3	5.6	96.41	96.12	0.00	0.00
10.93	66.2	28.5	103.98	103.68	0.02	0.00
11.93	95.9	58.2	111.55	111.25	0.04	0.00
12.93	110.6	72.9	119.12	118.82	0.05	0.01
13.93	278.7	241.0	126.69	126.39	0.17	0.02
14.93	417.8	380.1	134.26	133.96	0.27	0.03
15.96	2259.1	2221.4	149.43	149.14	1.58	0.17
16.20	3238.4	3200.7	152.98	152.68	2.28	0.25
17.00	4381.5	4343.8	164.80	164.50	3.10	0.33
17.26	4244.1	4206.4	168.64	168.35	3.00	0.32
18.10	4902.1	4864.4	181.06	180.76	3.47	0.37
19.30	5351.1	5313.4	198.79	198.50	3.79	0.41
19.96	7965.1	7927.4	208.54	208.25	5.65	0.61
20.96	9309.9	9272.2	223.32	223.03	6.61	0.71
21.96	9898.1	9860.4	238.10	237.81	7.03	0.76
22.96	10963.5	10925.8	252.88	252.58	7.74	0.83
23.98	11359.4	11321.7	267.95	267.66	8.03	0.86
24.96	11122.5	11084.8	282.44	282.14	7.86	0.84
25.96	11390.7	11353.0	297.22	296.92	8.05	0.86
27.96	12331.2	12293.5	326.77	326.48	8.71	0.94
28.94	12307.5	12269.8	341.26	340.96	8.70	0.93
29.96	13056.0	13018.3	356.33	356.03	9.23	0.99
30.96	12346.3	12308.6	371.11	370.81	8.72	0.94
31.92	12503.2	12465.5	385.30	385.00	8.84	0.95
33.09	11892.8	11855.1	402.59	402.29	8.40	0.90
33.94	11864.2	11826.5	415.15	414.85	8.38	0.90
34.93	12040.2	12002.5	429.78	429.48	8.51	0.91
35.96	12464.1	12426.4	444.63	444.33	8.81	0.95
36.96	12323.8	12286.1	459.05	458.75	8.71	0.94
37.96	12438.0	12400.3	473.46	473.17	8.79	0.94
39.06	12694.3	12656.6	489.32	489.03	8.97	0.96
39.97	13196.1	13158.4	502.45	502.15	9.33	1.00
40.95	13208.2	13170.5	516.57	516.28	9.34	1.00
41.96	13126.6	13088.9	531.14	530.84	9.28	1.00

SECTION D8
NICKEL EFFLUENT CONCENTRATIONS

Dates: 7/7/92 to 9/25/92

Column 4

Time (d)	Sample LSC (CPM/mL)	Sample - Background (CPM/mL)	Pore Volumes	Pore Vol. - Dead Vol.	Conc.	Relative Conc.
0.00	36.0	-1.7	0.06	-0.23	0.00	0.00
1.00	35.9	-1.8	15.14	14.84	0.00	0.00
2.00	43.1	5.4	30.28	29.98	0.00	0.00
3.00	38.5	0.8	45.42	45.12	0.00	0.00
4.00	34.8	-2.9	54.88	54.58	0.00	0.00
5.00	36.4	-1.3	62.45	62.15	0.00	0.00
6.00	32.3	-5.4	65.84	65.54	0.00	0.00
7.00	32.6	-5.1	73.41	73.11	0.00	0.00
7.93	36.9	-0.8	80.47	80.18	0.00	0.00
8.93	40.3	2.6	88.04	87.75	0.00	0.00
9.93	43.9	6.2	95.61	95.32	0.00	0.00
10.93	55.9	18.2	103.18	102.88	0.01	0.00
11.93	67.8	30.1	110.75	110.45	0.02	0.00
12.93	82.8	45.1	118.32	118.02	0.03	0.00
13.93	172.1	134.4	125.89	125.59	0.10	0.01
14.93	298.1	260.4	133.46	133.16	0.19	0.02
15.96	2290.6	2252.9	148.63	148.34	1.61	0.17
16.20	2873.3	2835.6	152.18	151.88	2.02	0.22
17.00	5279.6	5241.9	164.00	163.70	3.74	0.40
17.26	5003.9	4966.2	167.84	167.55	3.54	0.38
18.10	5849.2	5811.5	180.26	179.96	4.14	0.45
19.30	6384.2	6346.5	197.99	197.70	4.53	0.49
19.96	8966.9	8929.2	207.74	207.45	6.37	0.68
20.96	10016.3	9978.6	222.52	222.23	7.12	0.76
21.96	10205.9	10168.2	237.30	237.01	7.25	0.78
22.96	11301.2	11263.5	252.08	251.78	7.98	0.86
23.98	11630.2	11592.5	267.15	266.86	8.22	0.88
24.96	11383.1	11345.4	281.64	281.34	8.04	0.86
25.96	11441.1	11403.4	296.42	296.12	8.08	0.87
27.96	12479.4	12441.7	325.97	325.68	8.82	0.95
28.94	12307.5	12269.8	340.46	340.16	8.70	0.93
29.96	13142.1	13104.4	355.53	355.23	9.29	1.00
30.96	12310.7	12273.0	370.31	370.01	8.70	0.93
31.92	12472.7	12435.0	384.50	384.20	8.81	0.95
33.09	12107.5	12069.8	401.79	401.49	8.56	0.92
33.94	13205.8	13168.1	414.35	414.05	9.33	1.00
34.93	12679.0	12641.3	428.98	428.68	8.96	0.96
35.96	12509.7	12472.0	444.20	443.90	8.84	0.95
36.96	12398.5	12360.8	458.98	458.68	8.76	0.94
37.96	12490.6	12452.9	473.76	473.46	8.83	0.95
39.06	12863.9	12826.2	490.01	489.72	9.09	0.98

SECTION D8
NICKEL EFFLUENT CONCENTRATIONS

Dates: 7/7/92 to 9/25/92

Column 4

Time (d)	Sample LSC (CPM/mL)	Sample - Background (CPM/mL)	Pore Volumes	Pore Vol. - Dead Vol.	Conc.	Relative Conc.
39.97	13207.2	13169.5	503.13	502.84	9.34	1.00
40.95	13266.6	13228.9	517.26	516.97	9.38	1.01
41.96	13157.2	13119.5	531.83	531.53	9.30	1.00

SECTION D9
pH VALUES

INFLUENT SOLUTION:
Measured: 2/10/93

pH	Solution
7.31	10 mg/L Ni, 0.005 M CaCl2
7.22	10 mg/L Ni, 0.100 g/L NaN3, 0.005 M CaCl2

EFFLUENT SOLUTION:

	Sterile Columns		Inoculated Columns		daily	daily
Day	column1	column 2	column 3	column 4	average	st. dev.
3	8.18	7.85	7.97	7.9	7.98	0.15
10	7.88	7.96	7.92	7.9	7.92	0.03
17	7.22	7.26	7.7	7.71	7.47	0.27
24	7.78	7.76	7.58	7.51	7.66	0.13
49	7.27	7.29	-	-	7.28	0.01
54	7.36	8.03	-	-	7.70	0.47
average	7.62	7.69	7.79	7.76		
st. dev.	0.39	0.34	0.18	0.19		

	Column 1 and 2	Column 3 and 4
Average by column group:	7.65	7.77
Standard deviation:	0.35	0.17

overall average:	7.70
overall st. dev.:	0.29

Appendix E

Microbial Numbers

APPENDIX E

MICROBIAL NUMBERS

This appendix gives results of standard plate counts and direct microscopic counts. Results of plate counts from experiments 'V1' and 'L2' are summarized in Table 3 of Chapter 2. These plate counts were performed to determine the effect of Ni^{2+} in batch samples on culturable cell numbers for both freshly washed microbes (V1) and freeze-dried microbes (L2). Results of plate counts performed at the end of the columns experiment (C2) are given and are summarized in the 'Microbial Numbers' section of the Results and Discussion section of Chapter 2. This appendix also contains information from four direct counts which relate the optical density of various microbial suspensions to the number of cells. The direct count performed on the suspension used in experiment 'L2' was used to relate numbers of culturable cells to total cells.

PLATE COUNT RESULTS

Experiment: V1
NICKEL EFFECT ON THE VIABILITY OF FRESHLY WASHED MICROBES

FIRST COUNT:

	SPC A	SPC B	SPC C
	Plated: 2/29/92	Plated: 3/1/92	Plated: 3/2/92
	Counted: 3/4/92	Counted: 3/5/92	Counted: 3/6/92
Average (CFU/g tuff):	4.70E+07	1.07E+07	7.86E+06
St. Dev. (CFU/g tuff):	3.4E+06	1.6E+06	1.7E+06
n:	6	3	6

	SPC D	SPC E	SPC F
	Plated 3/2/92	Plated 3/2/92	Plated 3/2/92
	Counted: 3/6/92	Counted: 3/6/92	Counted: 3/6/92
Average (CFU/g tuff):	9.24E+06	1.03E+07	1.42E+07
St. Dev. (CFU/g tuff):	1.5E+06	3.8E+06	1.2E+07
n:	6	6	6

SECOND COUNT:

	Plated: 2/29/92	Plated: 3/1/92	Plated: 3/2/92
	Counted: 3/11/92	Counted: 3/12/92	Counted: 3/13/92
Average (CFU/g tuff):	4.66E+07	1.08E+07	7.91E+06
St. Dev. (CFU/g tuff):	3.0E+06	1.5E+06	1.7E+06
n:	6	3	6

	Plated 3/2/92	Plated 3/2/92	Plated 3/2/92
	Counted: 3/13/92	Counted: 3/13/92	Counted: 3/13/92
Average (CFU/g tuff):	9.27E+06	8.02E+07	1.33E+07
St. Dev. (CFU/g tuff):	2.4E+06	9.3E+07	1.2E+07
n:	3	6	6

SPC A = Sample take on the first day, prior to shaking.
SPC B = Sample take on the second day, after 24 hr shaking, prior to adding Ni.
SPC C = Sample taken on third day, no Ni solution added.
SPC D = Sample taken on third day, after shaking with .01 mg/L Ni for 24 hr.
SPC E = Sample taken on third day, after shaking with 1 mg/L Ni for 24 hr.
SPC F = Sample taken on third day, after shaking with 100 mg/L Ni for 24 hr.

PLATE COUNT RESULTS

Experiment: L2
NICKEL EFFECT ON FREEZE-DRIED MICROBE VIABILITY

	Plated: 9/16/92 Counted: 9/23/92	Plated: 9/17/92 Counted: 9/23/92	Plated: 9/18/92 Counted: 9/23/92
Average (CFU/g tuff):	2.91E+07	1.15E+07	1.19E+07
St. Dev.:	6.3E+06	1.4E+06	3.1E+06
n:	3	3	3
	Plated: 9/16/92 Counted: 10/8/92	Plated: 9/17/92 Counted: 10/8/92	Plated: 9/18/92 Counted: 10/8/92
Average (CFU/g tuff):	2.90E+07	1.13E+07	1.42E+07
St. Dev.:	6.1E+06	1.4E+06	2.3E+06
n:	3	3	3

Experiment: C2
CULTURABLE MICROBES IN COLUMNS AT THE END OF THE TRANSPORT EXPERIMENT

Column 1 and 2: Plated: 9/25/92
Counted: 10/8/92
All counts were <= 4 CFU/g; thus the number was statistically insignificant.
Counted colonies could have been from contamination of plates.

Column 3 and 4: Plated: 8/18/92
Counted: 8/25/92

	Column 3	Column 4
average (CFU/g tuff):	1.44E+05	1.73E+05
st dev (CFU/g tuff):	1.2E+04	1.3E+04
n:	3	3

DIRECT COUNTS

Experiment: no number
Date: 3/18/92
objective: Find the approximate number of 175 T#3 cells at an optical density of 0.8.
Average(cells/mL): 2.45E+08

Experiment: no number
Date: ?
objective: Find the approximate number of 175 T#3 cells at an optical density of 0.75.
Average(cells/mL): 3.76E+08

Experiment: L1
Date: 6/29/92
objective: Find the approximate number of 175 T3 cells at an optical density of 0.75.
Average(cells/mL): 5.14E+07

Experiment: L2
Date: 9/25/92
objective: Find the approximate number of 175 T3 cells at an optical density of 0.75.
Average(cells/mL): 5.22E+08
st dev: 1.42E+08
Note: The cells were difficult to count because they clumped together. For this reason, we doubled the original count to get these numbers.

Appendix F

Moisture Content Increase Resulting from Autoclaving

APPENDIX F

MOISTURE CONTENT ADDED BY AUTOCLAVING

This appendix gives information on the mass of water added to tuff by autoclaving. The mass of water present in the samples is given relative to the mass measured after the tuff was air dried for several days and before the first day of autoclaving. Note that water added by autoclaving evaporated, thus the mass of water in the sampes decreased from one day after autoclaving (A) to the next day before autoclaving (B).

The results show the mass of water added to the crushed tuff by the autoclave is negligable (<0.06 g)compared to the amount of solution added to samples for the batch sorption experiments (20 mL).

Experiment Date: Feb. 1992

Method

The experiment was done with triplicate samples. Sample averages are given in the results table.

1. ~10 g of crushed tuff was measured into polyallomer centrifuge tubes.
2. The tubes containing the tuff were weighed .
3. The tubes were autoclaved, then reweighed.
4. On days 2 and 3 the tubes were weighed again before and after autoclaving.

Results

Day	before/after autoclaving	ave. mass of water added (g)	st. dev. mass added
1	B	0	n/a
1	A	0.039	0.0034
2	B	0.008	0.0018
2	A	0.057	0.0034
3	B	0.011	0.0020
3	A	0.030	0.0072

This thesis is accepted on behalf of the faculty of the Institute by the following committee:

Robert A. Bowman 1 July 1993
Advisor Date

Thomas L. Kieft 7/1/93
Date

Hendricks 7/1/93
Date